Implementation of the Conservation Strategy for Tahoe Yellow Cress

(*Rorippa subumbellata*)

III. Pilot Project to Support Reintroduction Experiments

prepared for

Tahoe Yellow Cress Advisory Group

c/o Tahoe Regional Planning Agency

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Executive Summary

Here we report on the results of a pilot project designed to inform subsequent reintroduction experiments for Tahoe Yellow Cress (TYC), a plant endemic to the sandy shorelines of Lake Tahoe. Reintroduction is specified in the Conservation Strategy (Pavlik et al. 2002a) as a potentially valuable tool in efforts to restore and manage the species. The pilot project utilizes reintroduction to address critical gaps in our understanding of TYC restoration outlined in the "key management question" framework (Pavlik and O'Leary 2002). As we attempt to fill in these gaps, three goals guide our research: 1) Develop reintroduction logistics 2) Obtain basic data on TYC population biology and physiological ecology and 3) Obtain basic data on the effects of disturbance.

Beginning in May 2003 we worked with agency landowners to install the project at four sites around the southern and eastern perimeters of Lake Tahoe: Avalanche beach in Emerald Bay, Taylor Creek at Baldwin Beach, Zephyr Cove, and Sand Harbor. Each installation consisted of outplanting container-grown plants in "transect" configurations that extended from the waterline into different microhabitats. The number of transects installed depended on the width of the available beach, changes in lake elevation, human use patterns, archeological resources, and security. The transects were protected with fencing and signage and intrusions into fenced areas were monitored to see how people reacted to the installation. Demographic and water relations monitoring techniques were designed to assess microhabitat, genetic, hydrological, and recreational effects. We made site-specific predictions about TYC performance that could test our existing models and assist in designing future reintroduction efforts.

A total of 1,424 founders were installed across the four sites in May. By September, 815 founders were still alive. Among the sites, survivorship varied from a high of 85% at Avalanche beach to 58% at Taylor Creek and Zephyr Spit to low of 27% at Sand Harbor. Of the survivors, 58% were reproductive in September, producing an estimated 220,000 seeds (reproductive output was not calculated for Sand Harbor). The mean survivorship of all sites (57%) is slightly lower than that achieved in previous reintroduction efforts in 1988 (66%). Low lake levels persisted in both years, but it is likely that plants were not installed in the inundation zone in the previous projects. If the shoreline habitat is excluded then mean survivorship for the 2003 pilot project rises to a comparable 65%.

This pilot project was designed to meet a specific set of goals and objectives (pgs 9-10) and we can measure its success by those criteria. The three goals were prioritized such that logistical objectives trumped data gathering objectives for two reasons. First and foremost, the knowledge and ability to successfully produce and install TYC founders around the lake necessarily precedes any efforts to answer Key Management Questions. Secondly, previous outplanting efforts had shown varied levels of success but specific protocols were not developed to allow project duplication.

With regard to Goal 1 (develop reintroduction logistics), the pilot project proved to be a cost-efficient way to discover and solve logistical problems associated with propagating, transporting, and reintroducing a rare plant to its historical habitat. However, we failed to meet part of our first objective to propagate sufficient number of container-grown TYC founders for a full-scale experiment. In April 2002, the USFS contracted three nurseries to propagate TYC under greenhouse conditions but one of the nurseries did not produce any plants. By April 2003, only 1,665 plants were available for outplanting, just barely sufficient for the pilot scale outplanting and far short of the 4,000 plants required for a full scale experimental design. Many of the transplants varied in age by nearly four months, resulting in a set of founders that varied from small, vegetative individuals to plants that had gone to fruit in the greenhouse and were already beginning to senesce. A major finding of the pilot project was that founders with low initial vigor had significantly reduced chances of survival, emphasizing the importance of high quality founders. In the future we will want to track the source nursery of outplanted individuals to detect any difference in overall quality and minimize the effects on experimental reintroductions.

Proceeding with a smaller scale pilot design afforded us a great amount of success in developing protocols for site selection, outplanting, and monitoring and, consequently, we were able to meet many of the objectives of Goal 2 to obtain basic data on TYC biology. A key success was documenting the presence of an early season moisture gradient through the measuring of plant xylem water potentials. In June, water potentials of plants in shoreline were significantly higher (and therefore the plants were less water stressed) than plants in dune habitats. Interestingly, the gradient disappeared later in the season. In September, all sites experienced reduced survivorship in the shoreline microhabitat (<30% survival) compared to the high beach (>59% survival), but this was attributed to inundation and not water stress. This data will help us further refine our definitions of the various microhabitats so that we may better detect differences in survival and reproduction related to hydrology and microtopography.

Overall, the patterns of survivorship and reproduction among the four sites in 2003 are not easily explained by any single factor. The different seed sources showed differential survivorship within and among sites, but no clear patterns emerged that indicated any differential survival based on genetic factors. Other contributing factors include environmental variables such as microtopography, recreation impacts, and the influences of initial founder vigor. Some of these factors were controlled, but the lack of replication in the pilot design precluded efforts to determine the validity of observed trends and make robust comparisons for some variables.

Finally, we achieved success with the objectives of Goal 3 to obtain basic data on the effects of disturbance. Fencing effectively reduced human and other impacts and prevented disturbance-induced founder mortality. Installing access corridors through fenced plots appeared to further minimize disturbance and reduce the potential for vandalism, particularly during high visitation holidays (e.g. 4th of July). The success of the fencing in the pilot program provides a rationale for the installation and maintenance of fencing at future outplanting sites.

Introduction

The overall intent of the Conservation Strategy (CS) for Tahoe Yellow Cress (TYC, *Rorippa subumbellata*) is to restore a self-sustaining metapopulation dynamic that allows the species to persist in sandy beach habitat around Lake Tahoe despite high water levels and recreational impacts (Pavlik et al. 2002a). This report and two others (Pavlik et al. 2002b, Pavlik and O'Leary 2002) address preliminary work to advance Conservation Goals 2 and 4 of the CS. Goal 2 calls for improvement of the size and persistence of TYC populations at core and priority restoration sites. Goal 4 requires that research be conducted to directly support management and restoration activities.

A relevant and useful body of information about TYC biology, population persistence and associated habitat factors was summarized in the CS, providing an excellent opportunity for advancing the science of rare plant restoration. The monitoring database, covering more than 50 subpopulations over a 20 year period, is one of the most comprehensive available for any rare plant on earth. A solid understanding of TYC population dynamics was derived from that database, including a novel, empirically derived estimate of minimum viable population (MVP) size. This MVP estimate provides a target population size for creating new core populations around the lake (1,200 stems). In addition, several analyses of isozyme variability within and among subpopulations have provided the necessary basis for genetically composing the new populations. We also know that the species is a prolific seeder and can be readily cultivated under greenhouse conditions. Together, these factors make TYC an excellent candidate for a research driven reintroduction effort focused on filling information gaps necessary to restore the species.

Previous reintroductions were conducted at six sites in the late 1980's and early 1990's (CS p 19-20). Similar to 2003, these were years of drought and low lake level, and selection of outplanting sites was not based on hydrologic criteria. Five hundred plants each were planted at Meeks Bay, Tallac Creek, Baldwin Beach, and Taylor Creek and 1168 plants at D.L. Bliss in 1988. Survivorship in 1989 ranged between 33 and 93% (mean of 66% for all five sites). At Nevada/Kahle beach in 1991, 35 plants were salvaged and transplanted and 156 propagated plants were installed. This project met with very limited success, only 15 plants remained by 1994. These first attempts laid the groundwork for future restoration efforts by showing that it was possible to successfully propagate and transplant TYC.

However, neither of these projects contained a demographic monitoring component and there was no investigation of differential survival depending on a plant's original hydro-topographic position on the beach. Several specific objectives of the 2003 pilot project seek to address these gaps by focusing on demographic monitoring within different microhabitats.

The pilot project utilized small-scale, un-replicated reintroduction to address some of the practical gaps in our understanding of TYC restoration. It conforms to the "key management question" (KMQ) framework (Pavlik and O'Leary 2002) and primarily addressees the third KMQ, "Can TYC populations be created or enlarged in order to restore the self-sustaining dynamics of the species?" If we knew that TYC populations could be readily created in appropriate habitat, then land owners and managers would have a greater range of options available to them in designing and executing their projects. Although the question of how to restore TYC with reintroduction is complex and entails many stochastic variables, the pilot project goals address three main areas that affect reintroduction success: 1) reintroduction logistics 2) TYC population biology and physiological ecology and 3) disturbance.

Beginning in May of 2003, we worked with agency landowners to install the pilot project at four sites around the lake's southern and eastern perimeter. Three of the sites (Avalanche, Zephyr Cove and Sand Harbor) were used as "simple gradient" sites because they provided a single hydrologic gradient (measured as distance from the lake's waterline) and up to two microhabitats: moist shoreline and high beach. The other site (Taylor Creek) was used as a "complex gradient" site that provided several microhabitats (e.g. beach, beach trough, dune, meadow) with at least two hydrological gradients (e.g. distance from the lake's waterline and elevation above the water table). Approximately 1400 container-grown TYC individuals were outplanted among the four reintroduction sites.

During the 2004 field season we intend to implement a scientifically robust program of experimental reintroductions to more fully determine the habitat conditions and best management practices that optimize the chances for successful restoration of TYC. We will utilize a hypothesis-driven, replicated experimental design in the installation of several thousand container-grown plants at core and priority restoration sites around the lake. A replicated demographic monitoring component will enable us to best determine those factors that limit population growth and population persistence. Information from the pilot project on such factors as nursery propagation procedures, fencing, working with agency personnel, permit compliance, and outplanting and monitoring techniques will greatly inform the 2004 experiments.

Methods

Key Management Questions (KMQ's)

The KMQ's that guide conservation and restoration research on TYC (Pavlik and O'Leary 2002) are intended to implement the conservation strategy by focusing research on the restoration of metapopulation dynamics in the context of changing lake levels and continued human disturbances. Within an adaptive management framework, the KMQ's harness the power of a scientific approach while keeping the focus on generating information of immediate value to decision-making. The questions guide research to address specific, applied problems faced by land managers, agency regulators, and restoration biologists (Table 1).

Table 1. Key Management Questions

1) Can TYC populations occupy any site around the lake margin that has sandy beach habitat?
2) Are there ecosystem factors that can affect TYC performance within an occupied site or microhabitat?
3) Can TYC populations be crated or enlarged in order to restore the self-sustaining dynamics of the species?
4) Can any TYC genotype perform equally well at any appropriate site?
5) Can TYC microhabitats/places be found or created that are less likely to be adversely disturbed despite high visitor use or intense shoreline activity?

The 2003 pilot project primarily addressed the third KMQ, "Can TYC populations be created or enlarged in order to restore the self-sustaining dynamics of the species?" In an initial effort to answer this question, we looked at three main areas: 1) site factors that might determine the success of reintroduction or enhancement (e.g. microtopography, hydrology) 2) founder gene pool composition, and 3) logistical and security factors that might affect establishment (e.g. fencing). In a fully replicated experiment, we would have developed specific hypotheses to address each area. However, the pilot project was designed on a smaller scale to address initial objectives and goals (see below) and not as a replicated experiment that would provide statistical validation of hypotheses. Instead, the pilot project took full advantage of limited resources (funding and a small number of available founders) to investigate opportunities and barriers to creating or enhancing new TYC populations at four sites.

Pilot Project Goals and Objectives

The pilot project utilized small scale reintroduction to address several of the critical gaps in our understanding of TYC restoration outlined in the "key management question" framework (Pavlik and O'Leary 2002) As we attempted to fill in these gaps, the following goals guided our research: 1) Develop reintroduction logistics 2) Obtain basic data on TYC population biology and physiological ecology and 3) Obtain basic data on the effects of disturbance (Table 2).

Table 2. Goals and objectives of the 2003 Pilot Project.

G1: Develop reintroduction logistics

Ob 1: Nurseries: Develop nursery protocols to propagate container-grown, cold-hardened TYC founders

Ob 2: Site selection: Comply with all regulatory requirements to select suitable reintroduction sites

Ob 3: Outplanting: Develop protocols for planting TYC on different substrates

Ob 4: Monitoring: Develop monitoring protocols and datasheets, and train agency personnel

G2: Obtain basic data on TYC population biology and physiological ecology

Ob 5: Measure and compare survivorship and reproductive output in different microhabitats at 4 sites along east-west-gradient

Ob 6: Measure and compare plant xylem water potentials in different microhabitats

Ob 7: Track survival and reproduction of different genotypes in different microhabitats

G3: Obtain basic data on the effects of disturbance

Ob 8: Determine efficacy of different fencing and signage for minimizing human impacts

Ob 9: Evaluate benefits of installing access corridors through fenced plots (Taylor and Zephyr Spit)

Ob 10: Monitor exposure to human visitation (trampling, vandalism)

Ob 11: Determine potential for vandalism during high visitation holidays (e.g. 4th of July)

In lieu of a hypotheses-driven, replicated experimental design, we developed objectives for each goal to direct reintroduction efforts and maximize the amount of useful information we could obtain from the smaller scale project. Logistical objectives (Goal 1) focused on developing and refining protocols for propagating, transporting, outplanting, and monitoring TYC founders. These objectives met the need to standardize techniques and streamline procedures for subsequent experimental reintroductions, especially in light of an ever shifting composition of personnel on the TAG. A key part of the pilot project logistics was working with the TAG and engaging the agency land owners in the cooperative site selection process. Prior to outplanting it was necessary to obtain permits and comply with both federal and state regulations in order to install the pilot project (particularly fencing) on public properties with different regulatory oversight.

Data collection objectives (Goals 2 and 3) addressed TYC survivorship and reproduction in different microhabitats. Demographic, physiological, and disturbance monitoring techniques were designed to document responses of TYC founders to microhabitat, genetic, hydrological, and recreational factors. However, in the absences of replicated plots, the data can only suggest trends and functions mainly to inform subsequent experimental efforts.

Assignment of Tasks

We divided the pilot project into 32 discrete tasks (Table 3) in order to clearly designate the entity responsible for performing the work of the pilot project. Specifically, the tasks were divided between the agencies and private individuals that participate together as parts of the TYC TAG (e.g. U.S. Forest Service, California Department of Parks and Recreation, Nevada Division of State Parks, Tahoe Regional Planning Agency, etc.) and BMP Ecosciences (BMP). In the event that minor or unanticipated tasks arose that were not included in the table, they would be assigned by the following guidelines:

regulatory, logistic, field labor, and security tasks would be the responsibility of the TAG while training, design, technical, and archival tasks would be the responsibility of BMP.

Table 3. Tasks of the proposed pilot project, 2003-2004, and assignment of responsibility for those tasks.

	TAG	BMP
Plants		
1) Provide care for propagated TYC plants post-nursery	X	
2) Transport, pre- and post nursery	X	
3) Sort and color code by source, size, vigor	X	
4) Division into four equal founder groups	X	
5) Transport to outplant sites	X	
Sites		
6) Choose sites for outplanting	X	X
7) Evaluation and layout at each site, access & design	X	X
8) Fencing and signage, security	X	
Outplanting		
9) Assemble, inform and transport work crew	X	
10) Develop methods for outplanting		X
11) Train work crew at the sites		X
12) Assemble outplanting supplies and equipment		X
13) Supervise outplanting, quality control		X
14) Help crew collect microtopographic data		X
15) Final watering of founding plants	X	
Monitoring		
16) Post-outplanting evaluation @ mapping		X
17) Develop monitoring datasheets		X
18) Train monitoring crew		X
19) Monitor I - 10 day - supervise	X	X
20) Monitor II - 30 day	X	
21) Monitor III - 60 day	X	
22) Monitor IV - 90 day	X	
24) Monitor V - peak reproduction	X	
25) Data collation and transfer to BMP	X	
26) Water relations I		X
27) Water relations II		X
Data Analysis		
28) Data reduction		X
29) Data analysis		X
30) Graphical and tabular summary		X
Reporting		
31) Short report		X
32) Communication with TAG		X

Propagation of Founders

Seed Collection

BMP Ecosciences undertook the first steps of implementing the CS in the fall of 2001 with the field collection of TYC seeds. Seeds were collected in September at 9 priority and core restoration sites: Blackwood North, Blackwood South, Cascade, Edgewood, Lighthouse, Tallac Creek, Taylor Creek, Tahoe Meadows, and Upper Truckee East. We obtained seeds from a total of 177 individual plants across these nine sites (see Table 1 in Pavlik, Stanton, and Childs 2002). Collection protocols followed the Center for Plant Conservation guidelines for rare plants (Falk and Holsinger 1991). Over 46,000 seeds were carefully packaged and labeled for propagation at 3 nursery facilities. The entire 2001 seed lot was hand-sorted into three equal lots in December 2001 and stored in manila envelopes at room temperature and humidity until it could be delivered to the three nurseries. Each nursery received one third of the seed collected from each of the 177 individuals. These 177 seed lots each represented a specific seed source (site-based genotype) and individual plant. A small portion of seed was retained for laboratory germination tests.

Additional seed crops were collected from 11 sites in September 2002 and from 5 sites in September 2003. As part of the ongoing propagule production necessary for an age-structured reintroduction, the 2002 seeds were planted in July and August, 2003 (see below). The 2003 seeds are currently stored at room temperature and humidity in dry manila envelopes and will be sorted and planted in the summer of 2004.

2002 Nursery Propagation

Three nurseries received contracts through the USDA Forest Service to propagate TYC: the USDA Forest Service facility at an elevation of 3,200 ft in the Sierra foothills, just east of Placerville in Camino, CA; the USDA Forest Service facility at an elevation of 5,000 ft in Washoe Valley, NV; and privately-owned Sierra Valley Farms at an elevation of 5,000 ft in the Sierra Valley, 25 miles north of Truckee in Beckwourth, CA. Only the facility at Washoe had previously propagated TYC for restoration efforts. We chose to put the plants at three separate facilities to diffuse the risk of an unsuccessful propagation. Propagation protocols were developed in cooperation with these three nursery facilities to maximize yield of founding plants while minimizing artificial selection and *ex situ* loss of genetic variation. The objective was to raise hardy, rather than productive, founders that would survive transplanting. The research scheme calls for a sub-sample of founders to be held over to the next year for aging in anticipation of an age-structured outplanting.

The nurseries were directed to utilize all seed lots and plant a minimum of 1,400 plants in plastic supercells (Photo 1). Each seed lot represented an individual plant from a specific seed source. Nurseries planted as many as 20 replicates of a given seed lot and the actual numbers of planted supercells varied from 1,410 to 1,790. Detailed information associated with the propagules (e.g. seed lot, maternal parent ID) was tracked in order to estimate fitness components (e.g. seed output - plant size correlations) and evaluate the performance of different reintroduced populations. For further details see the first report in this series (Pavlik, Stanton, and Childs 2002).

2003 Nursery Propagation

The USDA Forest Service renewed contracts with two of the nurseries to propagate TYC for the anticipated 2004 reintroduction: the USDA Forest Service facility in Washoe Valley, NV, and privately-owned Sierra Valley Farms in Beckwourth, CA. Propagation efforts for over 4,000 TYC individuals began in July and early August of 2003. Established plants will be cold hardened in the late winter and spring of 2004 before they become available for the 2004 reintroduction.

Site Selection

Beginning in early 2003 we worked with agency landowners to identify four sites around Lake Tahoe that would be suitable for the pilot project. The four selected sites (Avalanche, Zephyr Spit, Taylor Creek, and Sand Harbor) are on lands belonging to three public agencies that are signatories of the CS. Selection of each site was based on a combination of the following factors: 1) the sites subjectively resemble "typical" TYC microhabitats, having the ecological characteristics described in the CS (pgs. 20-26) 2) the agency landowner could make an in-kind contribution of personnel for outplanting and monitoring 3) at high use sites, the agency could install fencing to protect the founders from human disturbance 4) the reintroduction and any associated fencing were feasible within CEQA or NEPA compliance, 5) the installation was compatible with the recreational patterns on the beach. In addition, it was desirable that the four selected sites span the west-east (mesic to xeric) microclimate gradients described in the CS (pg. 20).

Three of the sites (Avalanche, Zephyr Spit and Sand Harbor) were used as "simple gradient" sites because they provided a single microhabitat of "sandy beach" with one hydrologic gradient running perpendicular from the lakeshore towards stabilized vegetation. The other site (Taylor Creek) was used as a "complex gradient" site that provided several microhabitats (e.g. shoreline, beach trough, dune, meadow) with multiple hydrological gradients, including vertical and horizontal depth to the water table. In general, plot locations were chosen that subjectively resembled TYC habitat (naturally occurring plants may have been in the near vicinity) that also accommodated recreational use patterns. Descriptions of the individual sites are given below.

Taylor Creek (Complex Gradient)

The mouth of Taylor Creek (U.S. Forest Service), along the southwest shore of Lake Tahoe, is an important locality for TYC and is designated as a "Core" site in the CS. It has the highest viability index of any known TYC site (I = 97), largely due to high persistence (100%) and low coefficient of variation for mean maximum stem count (< 6%). Suitable habitat along this barrier beach has been available in high, low and transitional lake level years, which contributes to relatively constant subpopulation size (CS, Table 12). Overall habitat quality with respect to TYC is, therefore, demonstratively high, and we would expect high survivorship and reproductive output from founding individuals outplanted to optimal microhabitats. The beach itself, known as Baldwin Beach, stretches out to the west of the creek mouth and experiences heavy recreational impacts. Baldwin Beach has a viability index of –7 in the CS. Although plants have periodically been found on the main beach, the low index is likely due to the recreational pressure. We chose the extreme west end at the mouth of Taylor creek for the plot location because 1) several microhabitats were closely juxtaposed and could readily be fenced or had already been fenced because of TYC presence, 2) a pronounced hydrological gradient is produced by the creek-meadow-dune aquifer, and 3) the distance from the parking lot and heavily used portions of the beach would be a deterrent to opportunistic vandalism.

In May 2003 the following microhabitats were present near the Taylor Creek mouth: moist shoreline (exposed to wave action and inundation), beach trough (moist backshore depression, Knapp 1980), dune, dune trough (lagoon margin, Knapp 1980) and meadow. All of these could be contained within fenced areas (some temporary, some permanent fencing), while allowing for foot traffic to cross from the beach to the creek mouth and beyond. At the time of outplanting, a total of 48 naturally occurring TYC seedlings were found and flagged inside the fence and others appeared outside the fence in an east-west wrack line through the low beach microhabitat.

Based upon these known habitat features, we predicted that our pilot population at Taylor Creek 1) would have high survivorship and reproductive output in one or more microhabitat, and 2) overall survivorship would meet or exceed the mean survivorship (66%) observed in previous reintroduction efforts (CS, pgs. 19-20).

Avalanche (Simple Gradient)

Avalanche (California Department of Parks and Recreation) is to the southeast of Eagle Creek in Emerald Bay. The name derives from a rock avalanche that occurred above the site along Highway 89 in 1956. Avalanche beach is the small patch of readily inundated beach that is surrounded by logs swept down by that event. Avalanche beach itself is not ranked in the CS due to insufficient data. However, nearby Eagle Creek is ranked as a "High Priority Restoration Site" with a moderate viability index (I = 35) due to average persistence (56%) and moderate mean stem count and coefficient of variation (22%). At that site, suitable habitat has been available only in low lake level years (< 6,225 ft), which contributes to long gaps and intermittent patterns of persistence (CS, Figure 4). When available, overall habitat quality with respect to TYC is high, due to the shallow water table and relatively mesic microclimate (CS pg. 20). Similar site conditions exist at Avalanche and we would then expect high survivorship and reproductive output from founding individuals during low lake level years. The neighboring beach around the Vikingsholm property (north of Eagle Creek) is heavily visited, but recreational impacts to Avalanche have been light in the past. We chose the beach southeast of Eagle Creek for the plot location because 1) large, reproductive plants had been previously observed at this location (CSLC 1998, pg A-28) and were apparent later in the 2003 season, 2) the groundwater table would be just below the beach surface in this low lake level year and 3) the logs, creek and distance from the heavily used beach would restrict access and act as deterrents to opportunistic vandalism.

In June 2003 the two microhabitats available were moist shoreline and high beach. The tangle of downed logs on the beach and in the water deterred kayak beaching and foot traffic so no fence was installed. Signage indicating the beach closure was erected on the visited sides of the plot. At the time of outplanting, a small number of naturally occurring TYC seedlings were beginning to emerge.

Based upon these known habitat features, we predicted that our pilot population at Avalanche 1) would have high survivorship and reproductive output as long as lake level remained low, and 2) overall survivorship would meet or exceed the mean survivorship (66%) observed in previous outplanting efforts (CS, pgs. 19-20).

Zephyr Spit (Simple Gradient)

Zephyr Spit (U.S. Forest Service) lies just north of Zephyr Cove, Nevada on the east side of Lake Tahoe. Zephyr Cove is designated a "Medium Priority Restoration Site" in the CS, with a low viability index (I = 5) due to average persistence (50%), moderate mean stem count and high coefficient of variation (46%). Suitable habitat has been available in high, low and transitional lake level years (CS, Table 12). New subpopulations were found to the north at "Zephyr Spit" in 2001 and at "Zephyr Cove North" in 2002. Recreation pressures have likely driven the population away from the Cove toward these new sites on the less heavily used northern end of the beach. The location of these sites on the drier eastern shore of the lake (CS pg. 20) means a more xeric microclimate compared to Taylor Creek and Avalanche. We would then expect moderate to low survivorship and reproductive output from founding individuals. We chose the beach north of Zephyr Spit for the plot location because 1) it represents a site that can provide habitat regardless of lake level and 2) it could be readily fenced without blocking access.

In May 2003 the two microhabitats available were moist shoreline and high beach. Permanent fencing was installed on the upper reaches of the beach and a temporary fence was installed near the shoreline, in order to minimize recreational impacts. A footpath was available between the two enclosures. At the time of outplanting, naturally occurring TYC seedlings were present about 10m south of the fenced area and the cove population was emerging.

Based upon these known habitat features, we predicted that our pilot population at Zephyr Spit 1) would have low to moderate survivorship and reproductive output unless summer rains provided drought relief, and 2) overall survivorship would be less than the mean survivorship (66%) observed in previous outplanting efforts (CS, pgs. 19-20).

Sand Harbor (Simple Gradient)

Sand Harbor (Nevada Division of State Parks) is along the northeast shore of Lake Tahoe. It has been designated as a "Low Priority Restoration Site" by the CS because of its very low viability index (I = -38). Naturally occurring plants have not been seen here since 1979, resulting in low persistence (8%), low mean stem count and high coefficient of variation (47%). Suitable habitat may only be available in low or transitional lake level years (CS, Table 12), although sufficient data are lacking. Its location on the drier eastern shore of the lake (CS pg. 20) means a more xeric microclimate compared to Taylor Creek and Avalanche. We would then expect low survivorship and reproductive output from founding individuals. The beach, mostly located south of the boat ramps is heavily visited. Although Sand Harbor represents a site where the quality of habitat is probably lower than at more mesic sites, we chose the beach north of boat ramps for the plot location because 1) it could be readily fenced without blocking access and 2) Nevada Division of State Parks could provide monitoring personnel and quick compliance with state regulations.

In May 2003 the two microhabitats available were moist shoreline and high beach. A single fence was installed running perpendicular from the lake towards stabilized vegetation, leaving access open from the lake and no access corridor to the far north end of the beach. Large boulders and the slopes below highway 50 also provided some restricted access from the north and east. At the time of outplanting, no naturally occurring TYC seedlings were found.

Based upon these known habitat features, we predicted that our pilot population at Sand Harbor 1) would have low survivorship and reproductive output unless summer rains provided drought relief, and 2) overall survivorship would be less than the mean survivorship (66%) observed in previous outplanting efforts (CS, pgs. 19-20).

Design

Plant installations at the different sites consisted of outplanting container-grown plants in "transect" configurations that extended from the waterline into beach, dune, or meadow habitats (perpendicular to the shore). Establishing multiple plots was necessary at all sites to capture all of the microhabitat variation. Depending on the width of the available beach and changes in lake level, as many as 20 such transects were installed in a single plot. Transects were placed 1m apart and plants within a single transect were planted at 0.5 m intervals. Detailed planting protocols are in Appendix B.

The eight seed lots were distributed unequally across the four sites (Blackwood North and South were combined for outplanting). Three seed lots (Tahoe Meadows, Taylor Creek, and Upper Truckee) were planted at all 4 sites because these had demonstrated genetic variation in isozyme analysis (CS p14-17). The remaining seed lots were divided in half and put in two opposite lake quartiles. Within a site, a stratified random planting scheme was employed to distribute seed lots across the microtopographic gradient as evenly as possible. Plants were marked with a color-coded wire flag signifying the seed lot source.

Overall, the outplanting design was site-specific, lacking replication, and meant to address pilot project objectives rather than key management questions. The specific outplanting design for each site is discussed below. Site maps are in Appendix A.

Taylor Creek (Complex Gradient)

A total of 541 plants were installed in enclosures at Taylor Creek on May 19, 2003. This site had the most complex array of microhabitats and included: moist shoreline, beach trough, dune, dune trough, and meadow (high beach was not planted). The outplanting was divided into 5 plots, each representing at least one microhabitat. Plots 1 and 2, containing a total of 180 plants, were installed in the shoreline and beach trough area near the mouth of Taylor Creek and enclosed with temporary snow fencing (Photo 2). Throughout the season the creek inundated parts of plots, building up a berm along the moist shoreline, and creating a natural beach trough. Naturally occurring plants were beginning to emerge in Plot 2 and immediately outside the enclosure to the west. These were mapped and/or flagged and monitored through out the season.

In the higher beach, Plots 3-5 were enclosed with permanent fencing of wire and wood posts, adjacent to the existing enclosure that had been established by the USFS in the past. An inland lagoon (backbeach trough) bisected these enclosures and extended all the way down the beach toward the parking lot. Plots 3 and 4, containing 90 plants each, were situated around the margins of the lagoon to capture both dune trough, and dune habitats. An additional forty plants were planted behind the lagoon in stabilized vegetation in the meadow (Plot 5). A foot path 5m in width separated the shoreline plots from the plots on the high beach. Interpretative signage with a picture of TYC and accompanying information were placed on all sides of both enclosures.

In an effort to discern the effects of human trampling, forty plants were planted outside of the protection of the low beach enclosure. Ten plants each were planted within 10m of the fence on all four sides of the enclosure, including the footpath. Each plant was carefully mapped but not marked with a flag. There was no method to insure establishment of these plants, so it is not possible to strictly attribute the disappearance of any to trampling.

Avalanche (Simple Gradient)

Upper portions of this site were still covered with snow in mid-May, so planting was delayed two weeks to allow soils to dry. A total of 300 plants were installed on June 3, 2003 (Photos 3-5). Plants were arranged in 2 plots. Plot 1 contained 240 plants in 10 transects that extended out over a 12 m gradient from the waterline. These plants were in moist shoreline and high beach habitats. Exactly 60 plants were installed in Plot 2 in high beach habitat near 54 naturally occurring TYC plants that were just beginning to emerge. These natural plants were found above the plot near boulders and below the plot in a trough of beach wrack. The natural plants were mapped and monitored through the season. No fences were installed at this site because of its relatively remote location and the protection provided by downed logs from the avalanche. Plain signposts at the water's edge and on the western side of the plots indicated that the beach was closed for restoration.

Zephyr Spit (Simple Gradient)

The USFS provided two fenced areas on the beach just north of Zephyr Spit. Outplanting of 286 plants took place on May 22, 2003. Plot 1, enclosed with temporary snow fencing, contained 156 plants in moist shoreline and low beach microhabitats. Plot 2 abutted the bitterbrush/pine zone and was permanently fenced with wood posts and wire. It contained 130 plants, in high beach habitat. A footpath 5m in width separated the low beach plots from the high beach plots. The same interpretative signage used at Taylor was installed on the fences. Naturally occurring plants were found among the rocks out on the spit (30 m away) but these were not mapped or monitored.

Sand Harbor (Simple Gradient)

A total of 297 plants were installed on May 20, 2003. This site was very rocky, so it was necessary to work around boulders and divide the plants into 3 plots. Plots 1 and 2 each contained 120 plants and occupied both shoreline and high beach habitat. Plot 3 contained 80 plants, all within the moist shoreline habitat. On the southern end of the plots, adjacent to Plot 1, Nevada State Parks installed a snow style fence that extended from the lakeshore up into the stabilized bitterbrush/pine zone. Signage posted along the fence and at the waterline identified TYC but did not contain interpretative language. No naturally occurring plants have been found in the area.

Monitoring

Demographic, physiological, and disturbance monitoring techniques were designed to document responses of TYC to microhabitat, genetic, hydrological, and recreational factors. Detailed protocols are in Appendix B. A standard datasheet (Appendix C) was developed to record the fate of every outplanted individual, allowing subsequent calculations of mortality rates, survivorship to reproduction, and estimate reproductive output using models previously developed (Pavlik et al 2002b). The water relations monitoring component (Pavlik 1987, 2001), measured physiological stress levels (i.e. xylem water potentials) of plants established at different hydrotopographic positions with respect to lake level (datasheet also in Appendix C).

Demographic Monitoring

Each of the three agency landowners committed personnel for ongoing monitoring efforts throughout the 2003 growing season. BMP trained monitoring crews individually at each site (only once for both USFS sites) on the first scheduled monitoring day, two weeks after planting. Plants were evaluated at 2 weeks and 4 weeks after planting and thereafter on a monthly basis through October. Data collection parameters included: plant position, seed source, phenology, vigor, initial and final size, and current status. Initial plant size was measured during the 2-week monitoring and again in September at peak reproduction. Reproductive output was estimated based on an equation that links canopy size to seed output ($y=3.609x - 109.542$, $r = 0.81$) (see Figure 4, Pavlik, Stanton, and Childs, 2002).

Physiological Monitoring

Water relations monitoring was conducted twice during the 2003 growing season; once in early June, and again in late September during peak reproduction. An attempt was made to cluster the monitoring days and conduct the experiments under seasonally "typical" conditions: clear, sunny, warm, and not within 5 days after a storm front has passed.

Xylem water potentials were measured with a pressure bomb at two times during the day: predawn (5-6 am, before direct sunlight), and midday (2-4 pm), the period with warmest air temperatures and lowest humidity. If possible, two microhabitats were sampled at each site; moist shoreline, and high beach. Stems from a minimum of four TYC individuals were excised and immediately inserted into the pressure bomb for measurement. Within a microhabitat, individuals were selected based on position, apparent vigor, and sufficient size so that one stem could be excised without significant harm.

Disturbance Monitoring

Disturbance monitoring was conducted in conjunction with the demographic monitoring. An additional disturbance monitoring was conducted on July 7th in an attempt to document any impacts from the 4th of July weekend. At five times throughout the season, the monitoring crews made notes about the following possible disturbances in the plots: footprints/body impressions , animal prints (especially dogs and Canada geese), trash, and any acts of vandalism, especially those affecting TYC plants or the fence/signs. Photographs were taken of any significant disturbances and maps were generated to mark the areas of disturbance. Plot aisles and perimeters were raked smooth after all monitoring to obliterate any signs of disturbance and discourage people from entering the plots.

Results and Discussion

Founder Propagation

Two of the three nurseries successfully propagated TYC under greenhouse conditions. The outplanting at the USFS nursery in Camino did not produce any viable transplants, possibly as the result of heat stress from very high summer temperatures, coupled with over-watering. Sierra Valley Farms produced 1,048 and the USFS Washoe Valley nursery produced 1,103 plants. This represents greenhouse establishment rates of 64 and 778%, respectively (Table 4). Both nurseries kept approximately 250 plants to age for subsequent age-structured experimental plantings. In April 2003, a combined total of 1,665 plants were available for the pilot project. This number was just barely sufficient for the pilot scale outplanting and far short of the nearly 4,000 plants required for a full scale experimental design.

In early May, the plants were sorted at the Washoe Valley nursery according to seed lot and then assigned a vigor code (low, medium, and high) and a phenology code (vegetative, flowering, fruiting, and senescent). The vigor code partially reflected variability that resulted from different planting dates. Sierra Valley Farms planted their whole batch on May 21, 2002. While Washoe planted an initial group of less than 200 plants in late June that year, the majority of plants were not planted until early September. The fact that many of the transplants varied in age by nearly four months resulted in a set of founders that varied from small, vegetative individuals to plants that had gone to fruit in the greenhouse and were already beginning to senesce.

Of the 1,415 plants that were used in the outplanting, virtually none were in fruit at the time of planting, but 22% had flowered in the greenhouse and were classified as senescent. Of the remainder, 36% were vegetative and 41% were flowering. About 14% of the total had low vigor, while the rest of the plants were nearly equally divided into medium and high vigor (43 and 42%, respectively).

Table 4. Number of greenhouse-propagated TYC founders available for the 2003 pilot project in May, 2003 (0= not planted)

Seed Source	Sierra Valley Farms	USFS Washoe Valley
Blackwood North (BN)	13	8
Blackwood South (BS)	0	77
Cascade (CD)	0	102
Edgewood (ED)	31	37
Lighthouse (LT)	58	78
Tallac (TL)	109	55
Taylor Creek (TY)	156	201
Tahoe Meadows (TM)	143	163
Upper Truckee East (UTE)	293	129
Total for 2003	**803**	**850**
No. held over for 2004	**245**	**253**
Total established (ex situ)	**1048**	**1103**
Total planted (ex situ)	**1638**	**1410**
Percent established (ex situ)	**64%**	**78%**

The eight different seed lots varied with respect to initial vigor but no patterns were readily apparent (Figure 1). The most robust individuals came from the Tallac Creek (TL), Upper Truckee East (UTE), and Taylor Creek (TY). In contrast, Cascade (CD) and Blackwood (BS/BN) had very small proportions of high vigor founders and unusually high proportion of medium vigor plants. These two seed lots were among the earliest plantings at one greenhouse. Subsequently, many of the plants had fruited, were already beginning to senesce, and so were classified as medium vigor.

Demography of Founding Populations

Taylor Creek

Overall Performance: A total of 316 founders (58%) survived to the beginning of September at Taylor Creek. This is less then our prediction that overall survivorship would equal or exceed the previous transplanting effort (66%). However, survival in two microhabitats exceeded this level with 71% of the founders surviving in the beach trough and 73% surviving in the dune trough.

Of the surviving plants, 68% reached reproductive maturity and produced an estimated 119,085 seeds. In September, plant canopy size in reproductive individuals ranged from a low of 4 cm^2 (2 x 2 cm) to a high of 881cm^2 (40 x 27 cm), with the largest individual producing just over 3,000 seeds. Our method of determining seed output with a linear regression did not register positive seed output for any plants with a canopy less than 33 cm^2, although plants as small as 2 x 2 (4 cm^2) were observed bearing fruit. Within this huge range of variation, average plant canopy was 177 cm^2 (± 152 cm) and average seed output was 532 seeds/plant (±550 seeds/plant) (Table 5). Peak reproduction was observed through September and into October, with little or no decline in overall survivorship during this period.

Table 5. Reproductive output and total seed production at the four reintroduction sites, September 2003. (Sand Harbor was not evaluated).

Site	no. reproductive plants (#)	mean canopy size (cm2)	mean seed output (# per plant)	total seed production (# per site)
Avalanche	143	118.6 ± 88	318.3 ±318	44,794
Taylor	222	177.1 ± 152	532.5 ± 550	119,085
Zephyr	110	171.8 ± 145	510.7 ± 523	56,218
Sand Harbor	34	NA	NA	NA

Many individuals exhibited vigorous clonal growth during September (Photo 6). Small, vegetative "plantlets" appeared around 25% of the founders. At least 1,161 plantlets were counted, with a single individual founder producing anywhere from 1-45 plantlets. We only counted plantlets occurring within a 25 cm radius of a founder because this was the half way point between individuals in a transect. However, plantlets appeared at much greater distances. Vegetative reproduction was concentrated in the moist shoreline and beach trough microhabitats (Table 6).

In an attempt to identify connections between a plantlet and a "parent" plant, we excavated the plantlets of four individuals by carefully removing the soil from around the roots of the plantlet (Photo 7) . One individual had 4 plantlets growing at distances from 8 to 15 cm from the "parent" plant, but the roots of three were too fragile and broke before the connection to the parent could be positively established. We were able to identify positive connections between a single parent plant and 7 plantlets growing 4 to 15 cm from the parent plant (Photo 8). Additional attempts to excavate the root systems of plantlets growing at greater distances (>50 cm) from an identifiable "parent" plant were not successful because of the fragile roots. It is possible that some of these apparent "plantlets" were seedlings from new seed of the transplanted individuals. Experimental methods during the 2004 outplanting may be able to clarify the relative contributions of plantlet output and late-season seed germination to population growth.

Table 6. Plantlet output in different microhabitats at Taylor Creek, September 2003.

Site	(%) reproducing vegetatively	no. (#) of plantlets produced
shoreline	37	73
beach trough	43	1075
dune trough	3	3
dune	5	10
meadow	0	0
Total	**25**	**1161**

Founders placed outside the protection of the shoreline enclosure had low survival. Two weeks after planting, only 15 were present (37%). This is considerably lower than the 58% survival for the plants within the enclosures. All ten founders on the lakeside were inundated within a few days of planting and none reappeared later in the season. The founders nearest the creek mouth, in the area where natural TYC appeared in a line of beach wrack, fared the best. Although only 4 plants were present in July(10%), a few missing plants reappeared in August outside the fence, increasing the total number of surviving plants to 8 (20%). At this time, naturally occurring TYC individuals also appeared. It is impossible to pin point the reasons for the low survivorship of founders outside the fence. They may not have established upon transplant, they may have established, but gotten trampled, or they may have been inundated and washed away. A single plant within the 5 m path between the enclosures managed to persist through the season, possibly because a nearby shrub afforded some protection.

Effect of Microhabitat: Five microhabitats were present at the Taylor Creek site; moist shoreline, beach trough, dune, dune trough, and meadow, which supported plots 1-5, respectively. These habitats are characterized by large differences in hydrotopographic features such as lake inundation, wave action, wind erosion, depth to water table, or substrate deposition (see CS Table 2). Consequently, survivorship of TYC varied dramatically among the five microhabitats, ranging from 0% in the meadow to 73% in the dune trough (Figure 2). The high beach habitat, present at the other three outplanting sites, was not planted due to a shortage of founders.

Closest to the lake, the moist shoreline habitat was subject to periods of inundation during the early growing season. The lake continued to rise from spring run-off after planting in mid May and the mouth of Taylor Creek expanded into the low beach plot, forming both a berm in the moist shoreline habitat (plot 1) and a wet beach trough bisecting the middle of plot 2. Many founders were apparently washed away in the shoreline, reducing survivorship to only 28% by September. However, all of the surviving individuals were reproductive and these individuals were significantly larger in size and produced a higher number of seeds/plant than individuals in the other three microhabitats (Table 7). Survivorship apparently increased to 43% in October, as some plants probably re-sprouted after inundation. Although the "re-sprouts" appeared at or near the original planting point, they may have been new germinules and not from vegetative growth of the original founder.

Microhabitats further from the lake experienced less inundation and had higher survivorship of TYC founders than moist shoreline. Not surprisingly, survivorship was nearly equivalent in the two trough habitats (71 and 73% in beach and dune trough, respectively) (Figure 2). The proportion of individuals surviving to reproduction was also similar (73 and 64% in beach and dune trough, respectively). These microhabitats experienced mild inundation but also developed increasing amounts of vegetation (*Agrostis scabra*, *Juncus balticus*, *Rorippa curvisiliqua*) throughout the growing season. Plants in the drier dune habitat had 56% survivorship, while no founders in the dry meadow microhabitat survived past early July.

Table 7. Average canopy size and reproductive output in selected microhabitats at Taylor Creek, September 2003. (T- test significance level $p<.01$)

	# reproductive plants	avg canopy (cm2)	seed output/ plant	total seed production
moist shoreline	17	415.6 ±258.8 a	1390.3 ±934 a	23635
beach trough	129	160.4 ±105.2 b	469.3 ±379 b	60541
dune	34	197.9±168.2 b	604.7 ±607.2 b	20561
dune trough	47	114.9±117 b	305.3 ±422.3 b	14348

No clear patterns emerged regarding the performance of the six seed sources in different microhabitats. Founders from Upper Truckee East had nearly equal survivorship in beach trough and dune habitats

(Table 8). More variability was present in founders from the other seed sources, but without replication it is not possible to discern any significance.

Table 8. Percent survivorship of founders from different seed sources in selected microhabitats at Taylor Creek, September 2003. (* denotes insufficient sample size).

	Upper Truckee East	Tahoe Meadows	Taylor Creek	Cascade	Lighthouse	Tallac
moist shoreline	*	*	*	*	*	*
beach trough	70.3	66.7	77.8	63.9	69.6	68.8
dune	71.4	59.4	51.6	*	*	75
dune trough	*	93.3	100	*	87.5	53.3

Avalanche

Overall Performance: Compared to the other three reintroduction sites, survivorship of TYC founders was highest at the Avalanche site in Emerald Bay. Nearly 86% of founders (254 individuals) survived and 56% had survived to reproduce by September, greatly exceeding our predictions. Avalanche is exposed only in low lake level years and an increase in lake elevation of 2-3 ft would completely inundate the reintroduction site. However, during low lake level it appears that mesic conditions (shallow depth to the water table, shady, cool microclimate) offer optimal TYC habitat.

Total seed production for the site was estimated at 44,794 seeds. The average reproductive founder produced 318seeds/plant and had a canopy of 118 cm^2 (approximately 10 x 14 cm) (Table 5). Many individuals in the moist shoreline microhabitat exhibited vigorous clonal growth in September. Small, vegetative "plantlets" appeared around a total of 120 transplanted individuals. At least 868 plantlets were counted with a single founder producing anywhere from 1-34 plantlets. Peak reproduction was observed through September and into early October, with little or no decline in overall survivorship during this period.

Effect of Microhabitat: Two microhabitats were present at Avalanche; moist shoreline and high beach. The shoreline was subject to intense inundation and wave action early in the season and many of the founders planted within 2 m of the lakeshore were washed away soon after transplanting. Survivorship was only 17% by June, but in September it rose to 30%, possibly the result of re-sprouting of transplanted individuals or the emergence of new plantlets from surviving individuals (Figure 3). Of these survivors, 33% were reproductive, producing an estimated 1,290 seeds (n=4). Plants fared much better in the high beach microhabitat where they were not subject to inundation. As early as June, survivorship was almost 83%. High survivorship was sustained into September, barely decreasing to 82%. Almost 140 individuals were fruiting in September (57% of the survivors), producing an estimated 43,504 seeds (n=139). These reproductive individuals in the two microhabitats were not significantly different in canopy size or seed output.

Zephyr Spit

Overall Performance: Survivorship of TYC founders in the 2 plots (combined) at Zephyr Spit was 58% (167 individuals). We predicted low to moderate survivorship at this arid site in the absence of summer rains. It rained one week after outplanting but the amount of precipitation may not have been significant. Total seed production for the site was estimated at 56,218 seeds, with nearly 64% of surviving individuals reached reproductive maturity by September (Table 5). The average canopy size of reproductive individuals was 172 cm^2 and the average seed output was 510 seeds/plant. Many individuals exhibited vigorous clonal growth in September. A total of 63 founding individuals produced 894 plantlets, with a single individual producing anywhere from 1-50 plantlets.

Effect of Microhabitat: Two microhabitats were present at Zephyr Spit; moist shoreline and high beach. The shoreline was subject to protracted inundation and wave action while the lower reaches of the high beach habitat adjacent to the shoreline was only subject to mild wave action. Many of the plants along the shoreline were washed away soon after transplanting so that survivorship was only 6% in June, the lowest of any plot across all sites at the time. By September, survivorship had risen to 15% (Figure 4). Again, the re-emergence of plants was possibly the result of re-sprouting of transplanted individuals or the emergence of new plantlets from surviving individuals. None of the surviving individuals in the moist shoreline microhabitat reproduced in September.

Founders fared much better in the high beach microhabitat. Overall survivorship in September was 58% and the proportion surviving to reproduce was 67%. However, there was a pronounced difference in survivorship between plots 1 and 2. Individuals in the high beach portion of plot 1 had greater survivorship, were significantly larger and produced significantly more seeds per plant (Table 9). Plot 2 survivorship through out the growing season was quite low in comparison (Figure 4). Our definition of "high beach" lumps these two areas together, but one is clearly more suitable for TYC than the other. However, no significant difference was detected in measured water potential between plot 1 and plot 2 plants (see below). Zephyr Spit had a steeper gradient on the beach than either Avalanche or Sand Harbor, so our designation of "high beach" habitat may need refinement.

Table 9. Reproductive output in selected microhabitats at Zephyr Spit, September 2003.

	# reproductive individuals	Proportion reproductive	Mean canopy Size (cm2)	Mean # Seeds/plant	# plantlets
Moist shoreline	0	0	NA	0	0
Plot 1 beach	86	95%	205 ±146	633±527	888
Plot 2 beach	27	32%	48±31	65±112	6

Sand Harbor

<u>Overall Performance:</u> Survivorship of TYC founders was the lowest at Sand Harbor when compared to the other reintroduction sites. Sand Harbor was considerably drier and rockier than the other three sites and we predicted low survivorship, particularly in the absence of sufficient summer rain. Only 27% of founders (78 individuals) survived to September. Nearly all of plot 3 and a large portion of plot 2 were inundated and washed away early in the season. By September survivorship was 25% in plot 2 and less than 2% in plot 3 where all 5 rows experienced prolonged inundation and wave action. Although this site is arid, the data suggest that lake waters were the major cause of mortality at the site and not soil aridity. Plot 1 experienced less inundation because it contained over 100 plants that were arranged in 20 rows stretching away from the shoreline. September survivorship in Plot 1 was 44% and 48% of the surviving individuals were reproductive. Plant canopy size was not measured so no estimate of reproductive output is available for this site. Although new plantlet production was not recorded, very few individuals exhibited robust, clonal growth.

<u>Effect of Microhabitat:</u> Two microhabitats were present at Sand Harbor, moist shoreline and high beach. The shoreline was subject to intense inundation and wave action and many of the plants in the first 5 rows of all plots were washed away by July. June survivorship was 31% in the moist shoreline, but this decreased to less than 1% by August and it remained that way for the remainder of the season

(Figure 5). This is the only site that did not have plants re-emerge in the shoreline microhabitat and no reproduction occurred. In the high beach, survivorship was moderate at 63% in June, decreasing to 51% in September, and remaining around that level through October. Of these, 44% survived to reproduce.

Effects of Founder Initial Vigor

At Taylor, high vigor plants were three times more likely to survive than low vigor plants (79% compared to 26%, when the meadow microhabitat (plot 5) is excluded) (Figure 6). If low vigor plants managed to survive, they were actually more likely to reproduce than medium or high vigor plants (78% survivorship to reproduction, compared to 67%) (Figure 7). Similar to Taylor Creek, high vigor plants at Zephyr Spit were more than twice as likely to survive as low vigor plants and less likely to become reproductive. Although the differences were not as pronounced, the effects of founder initial vigor at Sand Harbor were very similar to those observed at Taylor Creek and Zephyr Cove. The smaller sample size of low vigor individuals overall may have contributed to their reduced survival, but it is more likely that low vigor founders simply succumbed to stress very early on. Consequently, a stress-induced hardiness may have developed in low vigor founders that made them more likely to reproduce if they survived.

Unlike the other three sites, initial founder vigor did not appear to influence survivorship at Avalanche. Low vigor plants were almost equally likely to survive as high vigor plants (81% compared to 89%) (Figure 6). This site did receive a smaller proportion of low vigor plants than the other sites (9% compared to 16 or 17%), but this marginal difference in plant allotments is probably not sufficient to explain the much greater overall survivorship at Avalanche. Interestingly, a greater proportion of surviving high vigor founders (65%) reproduced in September than low vigor founders (43%)(Figure 7). This reversal of the trends seen at the other sites may be directly related to site factors that lower stresses to the plants (i.e. more available water, less wind in a protected cove, an eastern aspect with less intense afternoon sun) but the contribution of these factors was not documented. Although not statistically validated, these trends highlight the importance of high quality plants to a successful reintroduction program.

Effects of Seed Source (Site-based Genotype)

A statistical analysis of the effect of seed source on survivorship and reproduction was not possible without replication, especially given the differences in initial vigor among the seed lots. At most sites, too few founders survived in the moist shoreline microhabitat to be able to detect any differences in survivorship resulting from seed source. Figure 8 shows survivorship in high beach microhabitat at all four sites for three seed lots (Tahoe Meadows, Taylor Creek, and Upper Truckee). These seed lots demonstrated genetic variation in isozyme analysis (CS p14-17) in previous studies (Saich and Hipkins 2000). At Avalanche, survivorship in the high beach was extremely uniform with over 90% for all three seed sources. At Zephyr Spit, survivorship in the high beach microhabitat of plot 1 was also very high (79-100%), but survivorship was more variable in the drier high beach habitat in plot 2. At Sand Harbor, seed from Upper Truckee East made a poor showing (only 20%) but there were only 15 plants from this source. In addition, the 50% survivorship seen in the Taylor Creek seed was based on only 2 founders so the small number of plants makes it difficult to draw any conclusions. Overall, no clear patterns emerged at any of the sites that indicated any differential survival based on genetic factors.

Demographic Comparisons Among Sites

A total of 1,424 founders were installed across the four sites in May. By September, 815 founders were still alive. Among the sites, survivorship varied from a high of 85% at Avalanche beach to 58% (Taylor Creek and Zephyr Spit) to a low of 27% at Sand Harbor. Of the survivors, 58% were reproductive in September, producing an estimated 220,000 seeds (reproductive output was not calculated for Sand Harbor). The mean survivorship of all sites (57%) is slightly lower than that achieved in previous reintroduction efforts in 1988 (66%). Low lake levels persisted in both years, but it is likely that plants were not installed in the inundation zone in the previous projects. If the shoreline habitat is excluded, mean survivorship for the 2003 pilot project rises to 65%. Despite the nearly perfect similarity in survivorship between the previous and current transplanting, the results are not strictly comparable. Many differences existed in outplanting design and monitoring and the important factor of initial founder vigor is virtually un-documented for the previous projects.

In the 2003 pilot, initial founder vigor appeared to have a strong influence on survivorship. Approximately 15% of the total founder population was classified as low vigor. At three of the sites mean survivorship of low vigor individuals was generally two and a half times less than that of high vigor individuals (26% compared to 65%). However, surviving low vigor plants were more likely to reproduce, possibly indicating a stress-induced hardiness. These trends were not apparent at Avalanche where low vigor plants faired just as well as high vigor founders and very high survivorship overcame the stress-induced effects of initial vigor.

Fencing effectively reduced the impact of variations in recreation intensity among the sites (see below). Avalanche had the highest survivorship, but it did not have any fencing installed because of its remote location. Sand Harbor had the lowest survivorship and the fence was often breached, but no plant mortality was attributed to human caused disturbance. Maintaining fencing throughout subsequent experimental plantings will be important for detecting other site-specific or genotype-related causes of differential founder survival.

Although some of the effects of natural environmental variables were not easily quantified, they likely played a major role in the differential survivorship of TYC founders among the reintroduction sites. All sites experienced reduced survivorship in the shoreline microhabitat (<30% survival) compared to the high beach (>59% survival), but this was attributed to inundation and not water stress. While the results suggest that high beach habitat is more suitable than the shoreline, without replication it is not possible to say that the shoreline habitats are equivalent or not. For instance, Taylor and Avalanche had nearly equal survivorship in the shoreline (28 and 30% respectively) in September, but all 17 of the survivors at Taylor were reproductive while only 33% of the 12 survivors at Avalanche developed fruit. The reasons for such differences are not clear.

Differences in water availability among the sites (see below), specifically the depth to the water table, could not be directly measured, but plant xylem water potentials offered an excellent surrogate measure. Average water potentials in June showed a significant hydrologic gradient running from the moist shoreline up into high beach or dune habitats at all sites. Water potentials of plants in shoreline were significantly higher (and therefore the plants were less water stressed) than plants in dune habitats. Low beach and high beach had intermediate levels that were also significantly different, indicating that water availability decreases as distance from the lake increases. However, these differences disappeared by September when there was no detectable gradient.

When comparing between the sites, the mean water potential of founders at Avalanche in September, the site with the greatest survivorship, were significantly higher than Zephyr Spit and Taylor, and therefore the plants experienced less water stress. It was anomalous, however, that Avalanche values were not as high as those at Sand Harbor, the site with the lowest survivorship. Mortality at Sand Harbor may not have been due to limited soil water availability, but further studies are needed.

The different seed sources showed differential survivorship within and among sites, but without replication it is impossible to demonstrate any significance to the observed performances. Differences in initial vigor between some seed sources appeared to explain some of the variation, but no clear patterns emerged that indicated any differential survival based on genetic factors.

Overall, the patterns of survivorship and reproduction among the four sites in 2003 are not easily explained by any single factor. Contributing factors include environmental variables such as microtopography (and depth to water table), recreation impacts, the influences of initial founder vigor, and the genetic source of the founders. Some of these factors were controlled, but the lack of replication in the pilot design precluded efforts to determine the validity of observed trends and make strict comparisons for some variables.

Water Relations of Founders

We measured the xylem water potential of TYC founders as a direct way to evaluate a plant's response to its immediate environment. Since plant water status reflects the ambient soil and atmospheric moisture conditions, it affords an opportunity to directly assess differences in the water availability among various TYC microhabitats. Well-hydrated plants have higher water potentials (less negative and closer to 0 MPa) because water is moving through the plant under low tension. As water becomes less available, plant water potential decrease (i.e. become more negative) and the plant experiences greater stress (e.g. loss of cellular turgor pressure). Water potential for forbs in mesic habitats generally ranges from at or near 0 MPa for a fully saturated plant to a lower threshold of –1.5 MPa for a sensitive plant that is stressed and near wilting.

The predawn water potential measurements had small variations between replicates and generally did not show any significant difference in plants from different microhabitats. The exception was at Taylor in June, where plants in the "beach trough" (plot 2) had significantly higher water potentials than plants in the "meadow" (plot 5) $p = 0.01$. This result was expected because the plants in the beach trough were in saturated soil conditions and some individuals were even submerged, while plants in the stabilized conditions of the meadow were much drier. These measurements were not repeated in September because all the plants in the meadow had died by early August.

Midday water potential measurements in June detected significant differences among plants within a site. Plants from the moist shoreline had significantly higher water potentials (and therefore less water stress) than those in other microhabitats.(Figure 9). Plants in the true "shoreline" (only measured at Sand Harbor) had an average midday water potential of -0.1 MPa while plants in the high beach at Zephyr Cove and Sand Harbor had midday water potentials of –1.3 and –1.4 MPa, respectively. Founders in the dune microhabitat at Taylor had the lowest average midday water potential of –1.7 MPa. These data suggest there was an early season gradient of increasing water stress at midday from moist shoreline to high beach and into the dunes.

This gradient of water stress between microhabitats completely disappeared by September. Within a site, plant water potentials did not differ significantly, indicating that the surviving individuals were experiencing similar levels of water stress (figure 9). Among sites, Avalanche founders had significantly higher water potentials than those at Taylor and Zephyr Cove (but not Sand Harbor). The higher water potentials at Avalanche may partially explain the higher survivorship of founders (85%). However, the fact that water potentials were not significantly different from Sand Harbor, where survivorship was only 27%, indicates that water potential may not be the only factor significantly affecting the population.

One goal of the 2004 experiment will be to use water relations data to further define microhabitats and make comparisons of plant performance in a single microhabitat that occurs at different sites. Microhabitats were not objectively defined at the beginning of the 2003 pilot project, so strict comparisons between and among the sites were not always possible or defensible.

Effects of Disturbance

Exposure to Typical Summer Recreation

The four reintroduction sites experience a range of recreational pressures, from light foot traffic to large volumes of people and pets. In general, recreational use is highest in July and August and more moderate in June and September. The beach at Avalanche gets the least use, even though Emerald Bay is one of the most visited places in the Tahoe Basin. The beach itself is difficult to access and receives very light foot traffic, mostly from people in kayaks or boats. At Baldwin Beach, the USFS charges $5 to park during the summer and the beach can get very crowded on the weekends. However, the Taylor Creek enclosures are at the far eastern end of the beach and lightly visited. Zephyr Cove is operated by an independent company that charges $7 to park at the resort, which includes volley ball pits on the main beach, a bar, and a restaurant. It is very crowded throughout the peak summer months, but the enclosures at Zephyr Spit are on the north end of the beach, furthest from the parking lot. Sand Harbor is the largest beach on the east shore and also receives heavy use all summer long. It costs $7 to park at the boat beach, but parking is limited to vehicles transporting boats or kayaks. Although the boat beach is small, it gets overflow from the main beach and wave action can be very high from jetski and boat traffic. Dogs are not allowed at Avalanche or Sand Harbor, but dogs were occasionally to frequently spotted on all beaches. While dogs cannot get through the temporary snow fencing, they can easily get through the wire permanent fencing.

None of the enclosures were vandalized during the course of the pilot project. California State Parks reported on one occasion the presence of human and bear tracks in the plots with no corresponding damage (intentional or otherwise). At Taylor, a set of footprints was found inside the temporary snow fence, but no damage was reported. Some of the flags were pulled out of the temporary enclosure at Zephyr Spit, probably from people reaching over the fence, and the fence was also cut on one occasion. Sand Harbor had the greatest amount of incursions into the planted areas as people walked through the upper part of the plots and along the water on a regular basis (accessing the area directly from the water) but there were no reports of any human-caused loss of founders. Many of the flags were inundated and people apparently gathered up the stray flags, but there was also trash occasionally scattered through the plots.

Fourth of July Holiday

No vandalism occurred on the Fourth of July holiday weekend, a period of extremely high visitation. Local observers had noted in the past that populations of TYC suffered light to heavy trampling damage during the period. Although the USFS beaches and Sand Harbor were at maximum capacity, no damage to fencing or transplanted individuals was reported during the July 7thdisturbance monitoring.

Fencing, Access Corridors and Signage

Fencing was very effective at protecting the reintroduced populations at Taylor Creek and Zephyr Cove. The access corridors between the plots at both sites allowed people to move past the plots without any disturbance and the signage on the fencing had a photo of Tahoe Yellow Cress and a small amount of information about the species. On one monitoring occasion at Zephyr Cove, a boy was overheard telling his companion to watch out for the "electric fence", so a certain intimidation factor may have played a role in people respecting the plot boundary. It may also be that leaving the tall wire flagging in place near each plant helped people to see there was a project going on and not just a collection of random plants.

The fencing at Sand Harbor was much less successful in keeping people out of the plots. The lack of any access corridor meant that people had to either find the path above the plots or walk into the water in order to get to the big rocks at the end of the cove. Another factor contributing to people entering the plots may have been the lack of a picture and interpretive information on the signs explaining the nature and value of the project.

No fencing was installed at Avalanche because of the remote location and difficult access. Signs were not project specific and only instructed people to "keep out of plant rehabilitation area". However, the flagging clearly showed that a project was ongoing and the few visitors to the beach appeared to respect the restriction.

Reintroduction Logistics

The pilot project proved to be a cost-efficient way to discover and solve logistical problems associated with propagating, transporting, and reintroducing a rare plant to its historical habitat. The first problem occurred when collected TYC seeds failed to germinate in laboratory tests, delaying seed sowing at Washoe Valley and Camino (see Pavlik, Stanton and Childs 2002). Sierra Valley Farms was unaware of the delays and had already planted their seed lots by May 2002. The next setback occurred in fall when it became apparent that plants at Camino were not surviving cultivation at low elevation. It was fortunate that we spread the propagation out over three nurseries to reduce the risk of ending up with no plants for subsequent reintroduction. However, the different planting times made for an extremely variable founder population and this variability confounded efforts to determine the effects of environmental and genetic parameters on plant and population performance. Ideally, only one nursery would produce plants in order to decrease variability of size and vigor in the founders.

The USFS and BMP transported the plants from the Washoe Nursery to the USFS workstation in Meyers the week before outplanting. During the sorting process, the plants from Sierra Valley Farms and Washoe were mixed together so it was not possible to tell if one nursery produced superior quality plants. This information would help select a single nursery for future propagations.

The USFS and NDSP installed the fencing at the outplanting sites in a timely manner. When the outplanting began, the actual planting process went fairly smoothly and there were sufficient personnel to help at each of the four sites. Rocks were moved if possible, or the space was left unplanted and mapped as such. About 25 founders were not outplanted in an area around the lagoon margin at Taylor Creek because of archeological concerns, but this did not significantly alter the overall design. Planting at Avalanche was delayed two weeks because of snow and it would be preferable to have all sites planted at the same time.

Conclusions

This pilot project was designed to meet a specific set of goals and objectives (pg 3) and we can measure its success by those criteria. The three goals were prioritized such that logistical objectives were considered more important than data gathering objectives for two reasons. First and foremost, the knowledge and ability to successfully produce and install TYC founders around the lake necessarily precedes any efforts to answer Key Management Questions. Secondly, previous outplanting efforts had shown varied levels of success but specific protocols were not developed to allow project duplication.

With regard to Goal 1 (Develop reintroduction logistics), the pilot project proved to be a cost-efficient way to discover and solve logistical problems associated with propagating, transporting, and reintroducing a rare plant to its historical habitat. However, we failed to meet part of our first objective to propagate sufficient number of container-grown TYC founders for a full-scale experiment. The plants that were available for the outplanting were extremely variable in terms of age and vigor and this hindered efforts to determine the origins of differential survivorship and reproduction. A major finding of the pilot project was that founders with low initial vigor had significantly reduced chances of survival, emphasizing the importance of high quality founders. In the future we will want to track the source nursery of outplanted individuals to detect any difference in overall quality and minimize its effect on the reintroduction experiment

Proceeding with a smaller scale pilot design afforded us a great amount of success in developing protocols for site selection, outplanting, and monitoring and, consequently, we were able to meet Goal 2 to obtain basic data on TYC biology. A key success was documenting the presence of an early season moisture gradient through the measuring of plant xylem water potentials. These data will help us further refine our definitions of the various microhabitats so that we may better detect differences in survival and reproduction related to microtopography.

Finally, we achieved success in meeting objectives of Goal 3 to obtain basic data on the effects of disturbance. Fencing effectively reduced human and other impacts and prevented disturbance-induced founder mortality. Installing access corridors through fenced plots appeared to further minimize disturbance and reduce the potential for vandalism, particularly during high visitation holidays (e.g. 4th of July). The success of the fencing in the pilot program provides a rationale for the installation and maintenance of fencing at future outplanting sites.

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Thank you to Kim and Garry Romano at Sierra Valley Farms and Rick Viggen and Casey at the USFS Washoe Valley Nursery for the successful propagation of a founding cohort of TYC. The USFS LTBMU provided funding for the nursery propagation effort and fencing materials.

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Figures

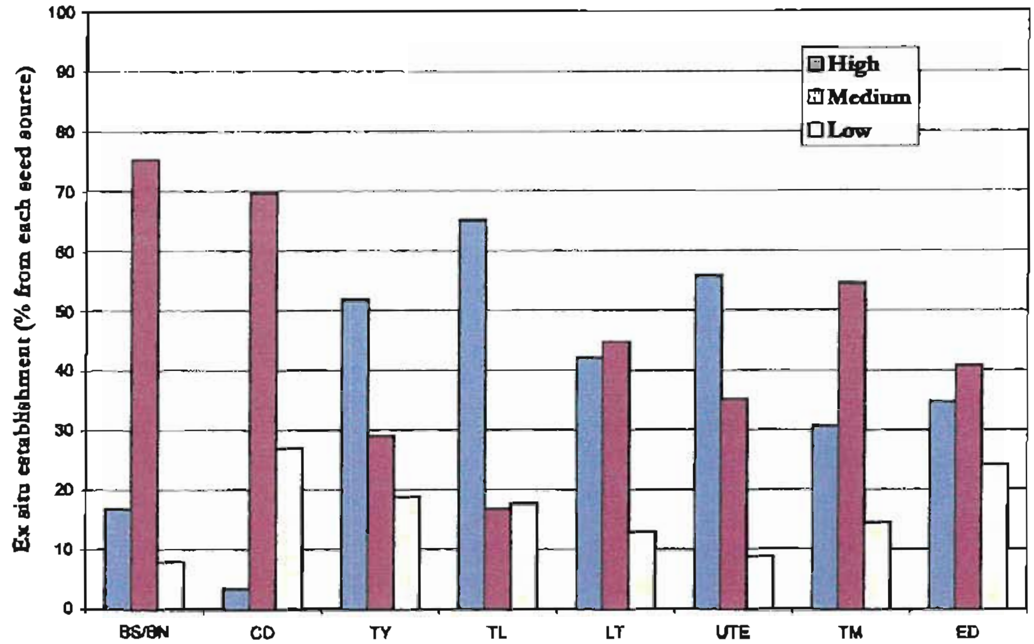


Figure 1. Initial vigor of TYC founders from different seed sources in the 2003 greenhouse propagation.

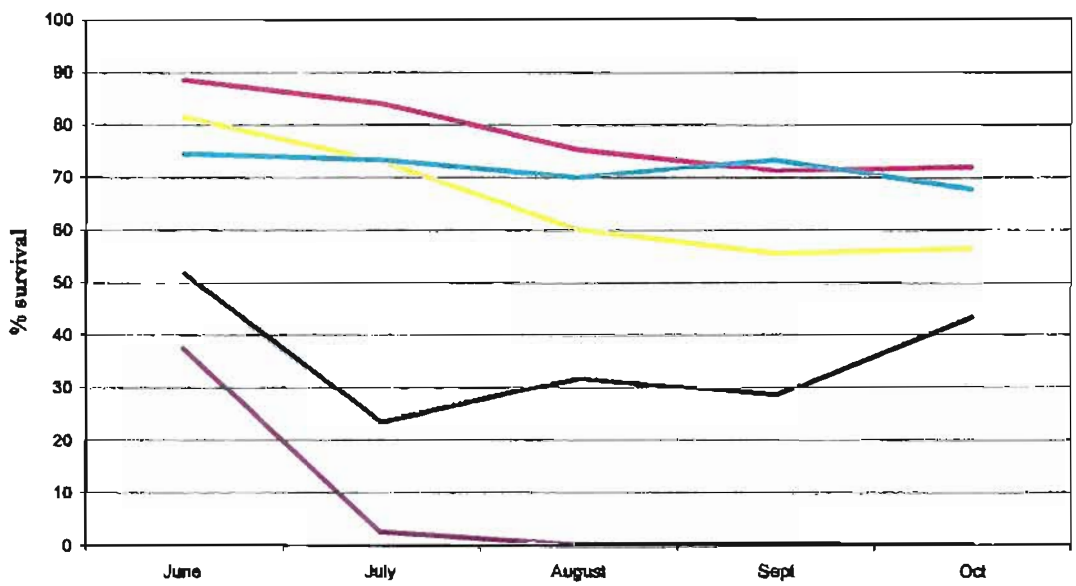


Figure 2. Survivorship in different microhabitats at Taylor Creek, June- October 2003.

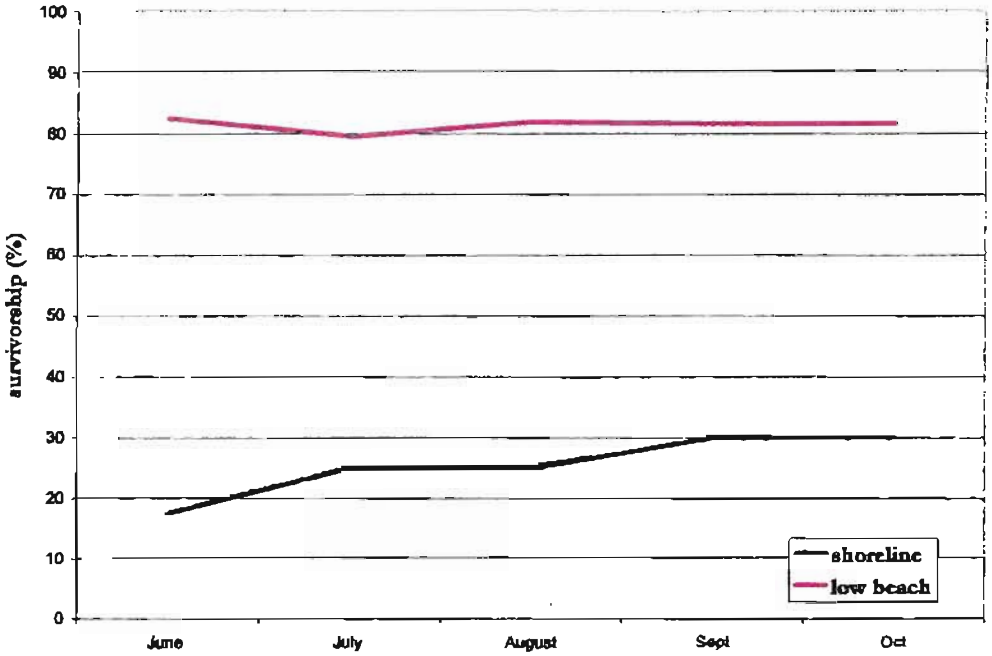


Figure 3. Survivorship in two microhabitats at Avalanche, June-October 2003

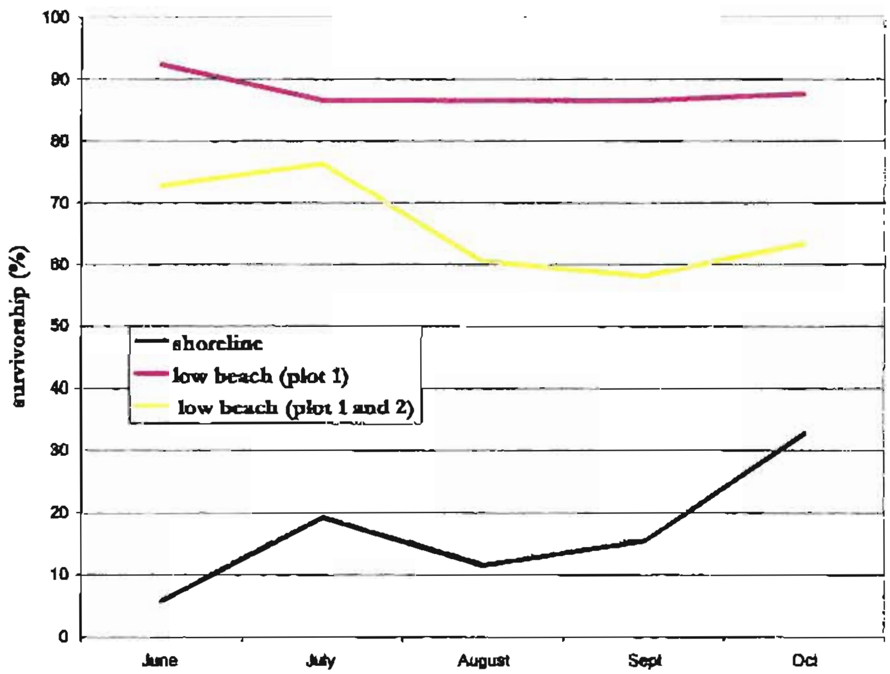


Figure 4. Survivorship in three microhabitats at Zephyr Cove, June- October 2003.

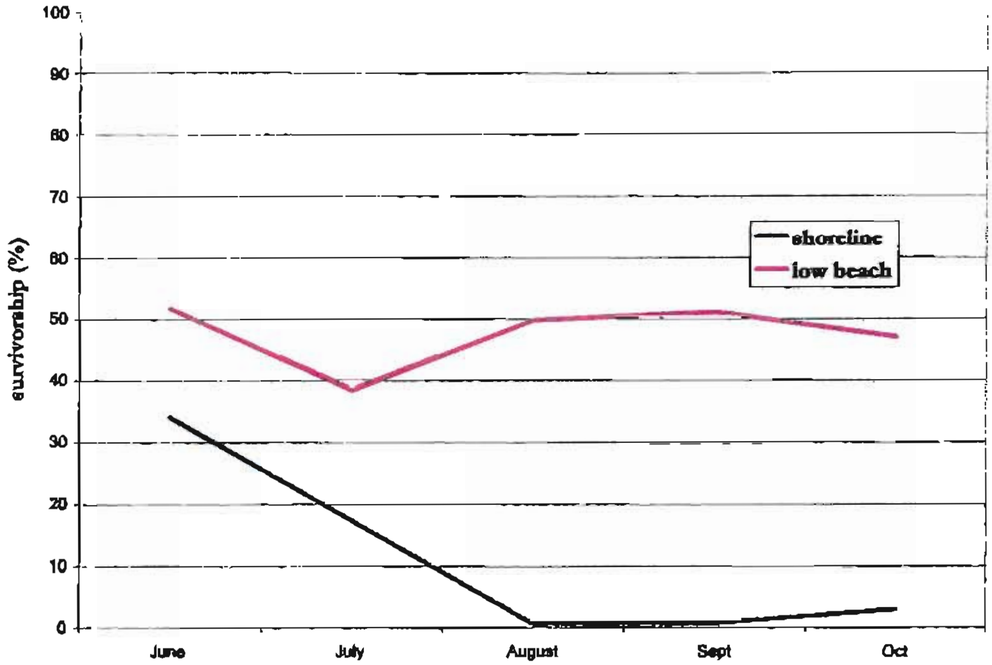


Figure 5. Survivorship in two microhabitats at Sand Harbor, June-October 2003.

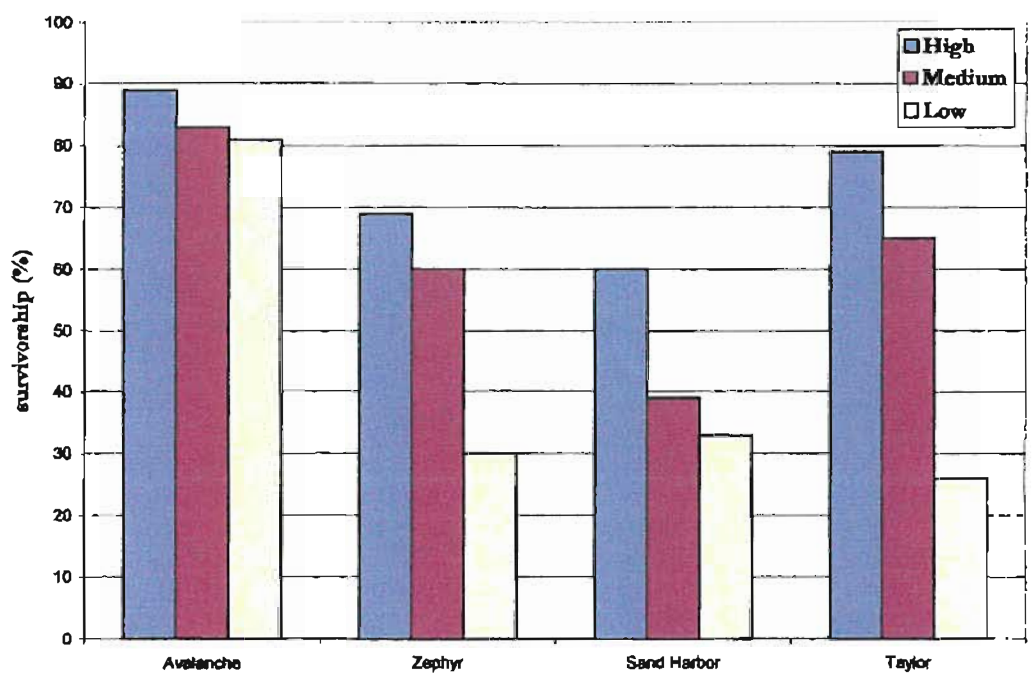


Figure 6. The effect of initial founder vigor on survival at all four sites, September 2003.

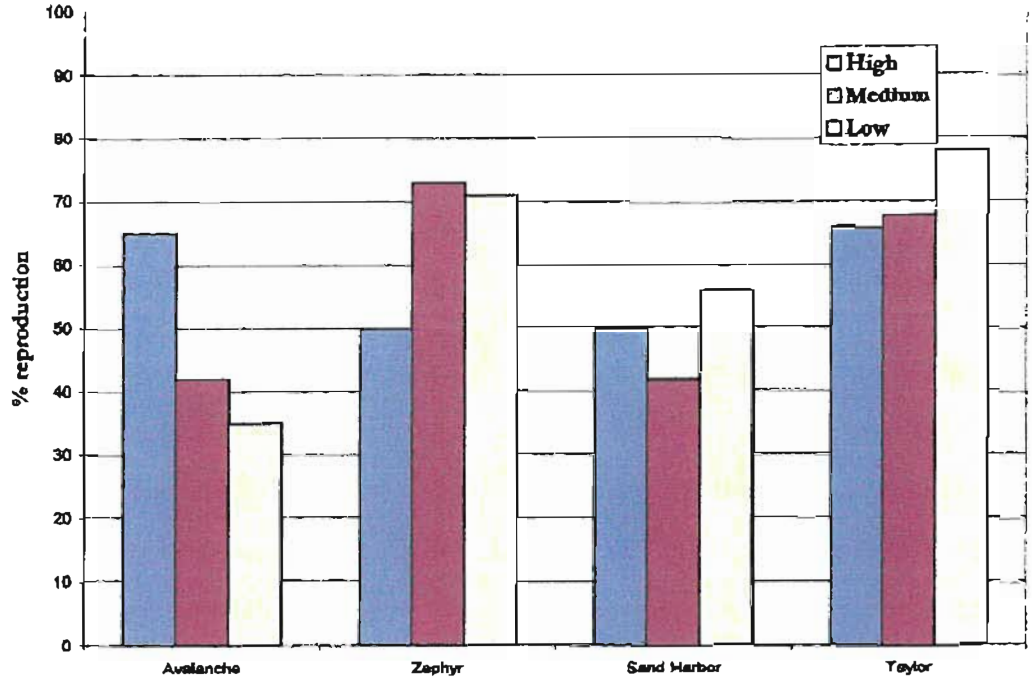


Figure 7. The effect of initial founder vigor on survivorship to reproduction at all sites, September 2003.

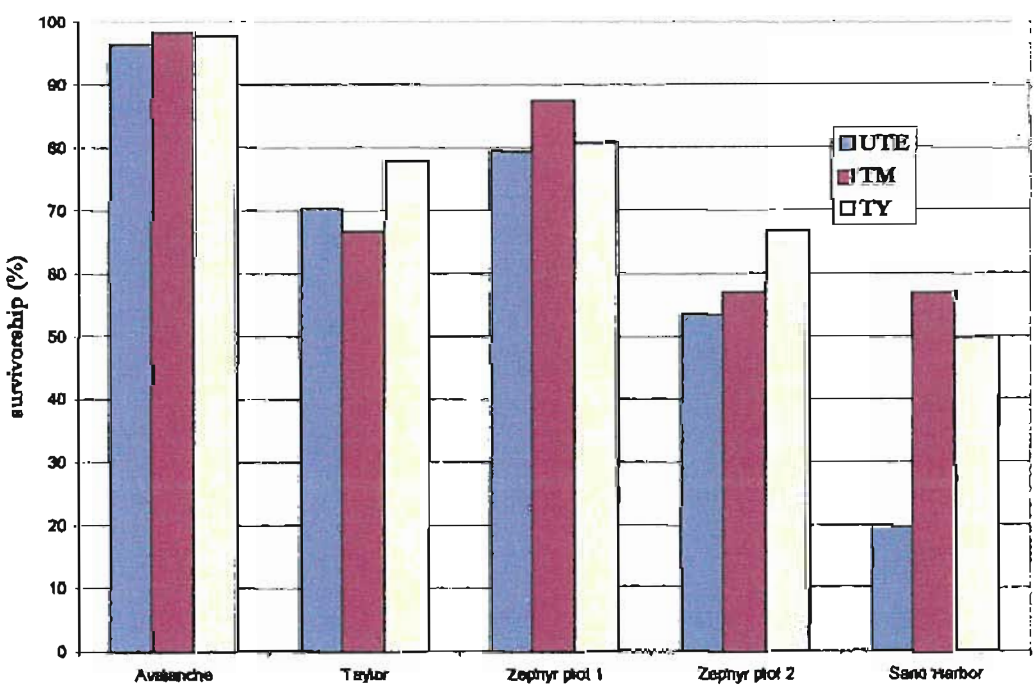


Figure 8. Survivorship (%) of selected seed sources in low beach microhabitat, September 2003.

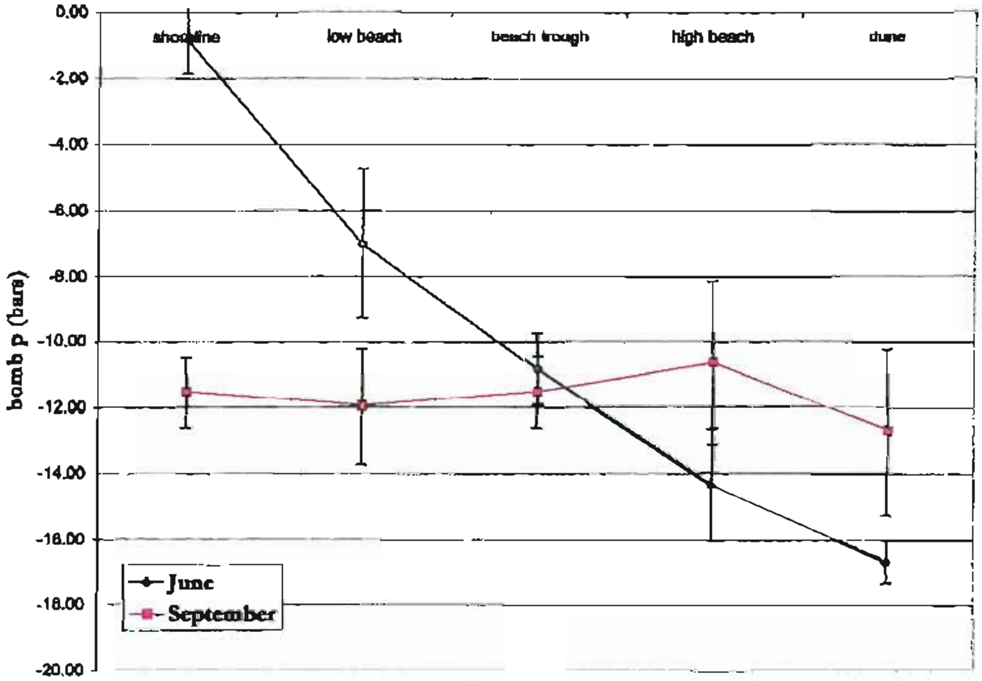


Figure 9. Mid-day water potentials of TYC in selected microhabitats, 2003.

Photo 1. Container-grown TYC in the Sierra Valley Farms greenhouse, August 2002.

Photo 2. Plot 1 shoreline and Plot 2 beach trough microhabitat at Taylor Creek.

Photo 3. CDPR crew installing TYC at Avalanche beach, June 2003.

Photo 4. Inserting TYC plant in the ground at Avalanche beach.

Photo 5. Container-grown TYC showing root stucture

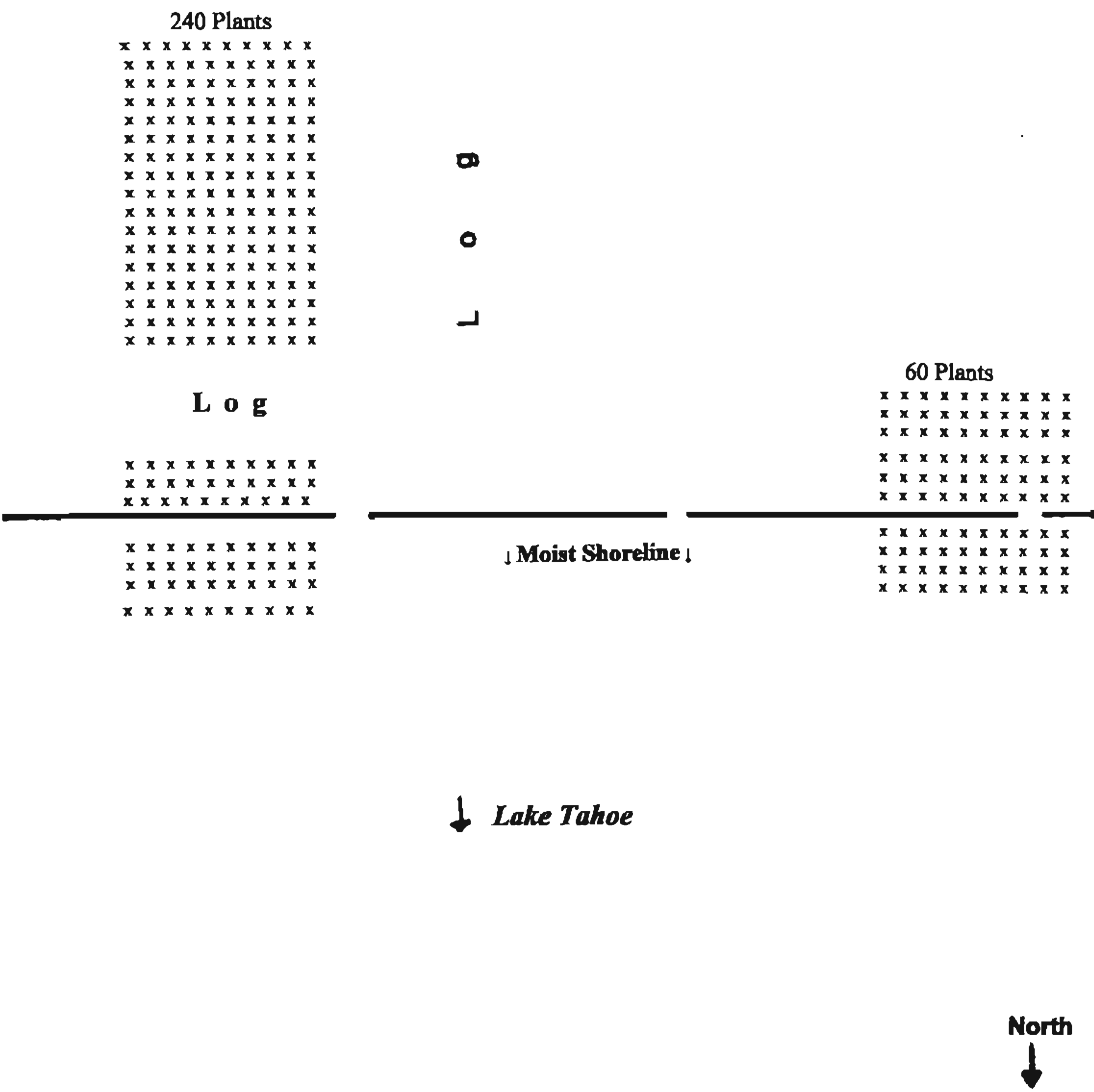
Photo 6. New "plantlets" appearing a round a TYC founder in September.

Photo 7. Excavated TYC plant with exposed root system at Taylor Creek.

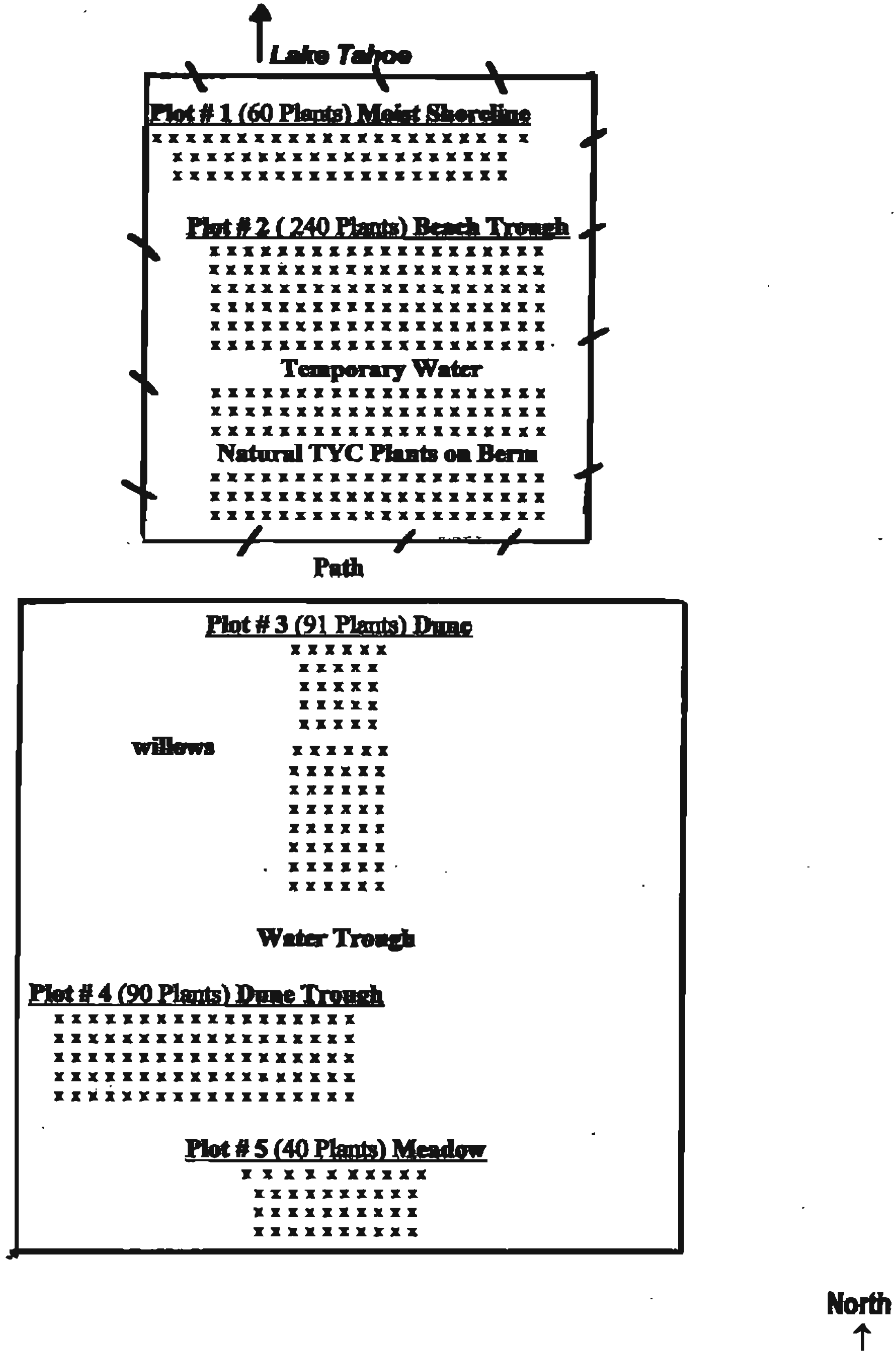
Photo 8. TYC root network showing the underground connections of "plantlets".

Appendix A. Site Plot Maps

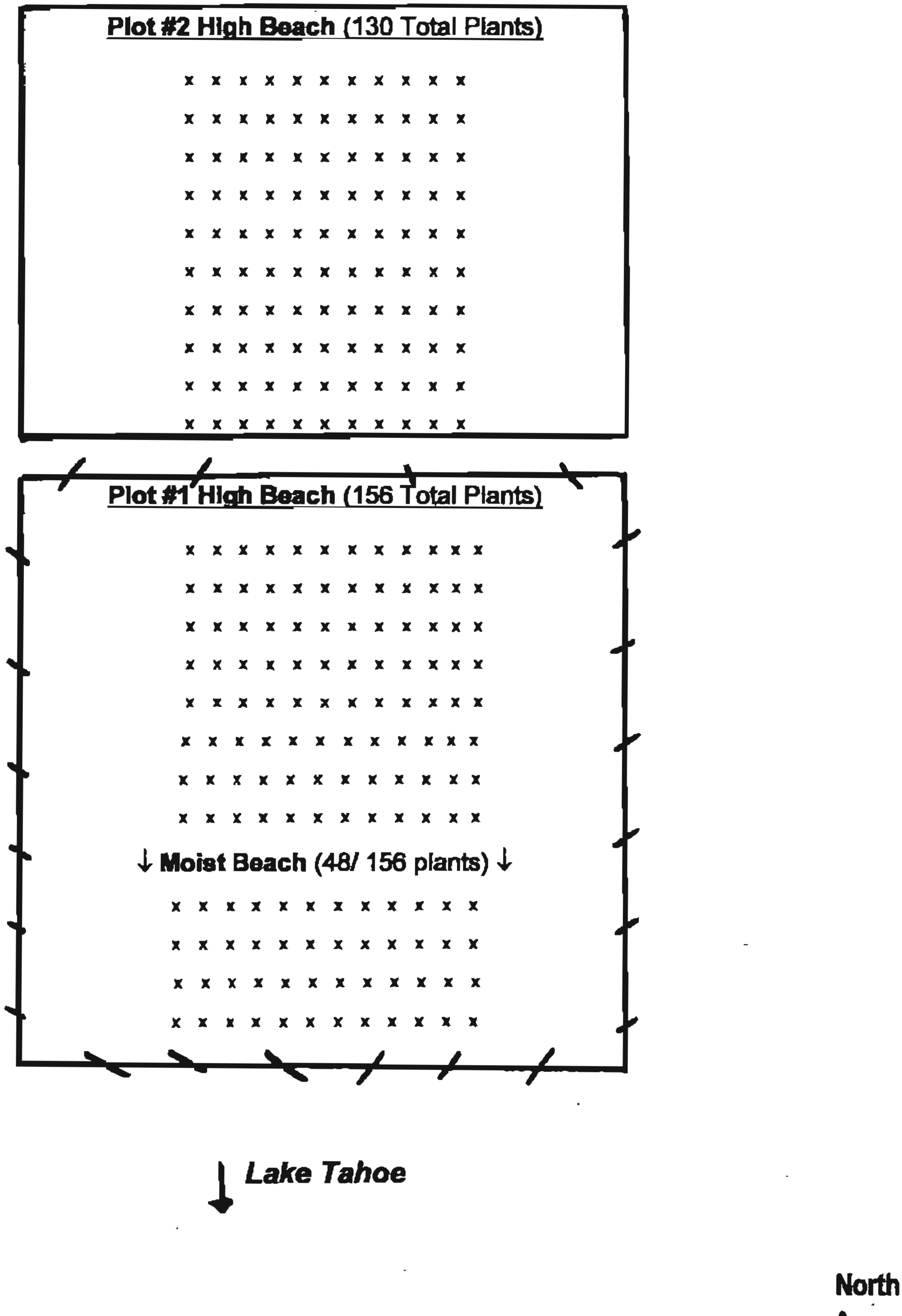
A1. Avalanche Beach, Emerald Bay (CDPR)
Site Map 2003 Pilot Project
(Total Plants = 304)



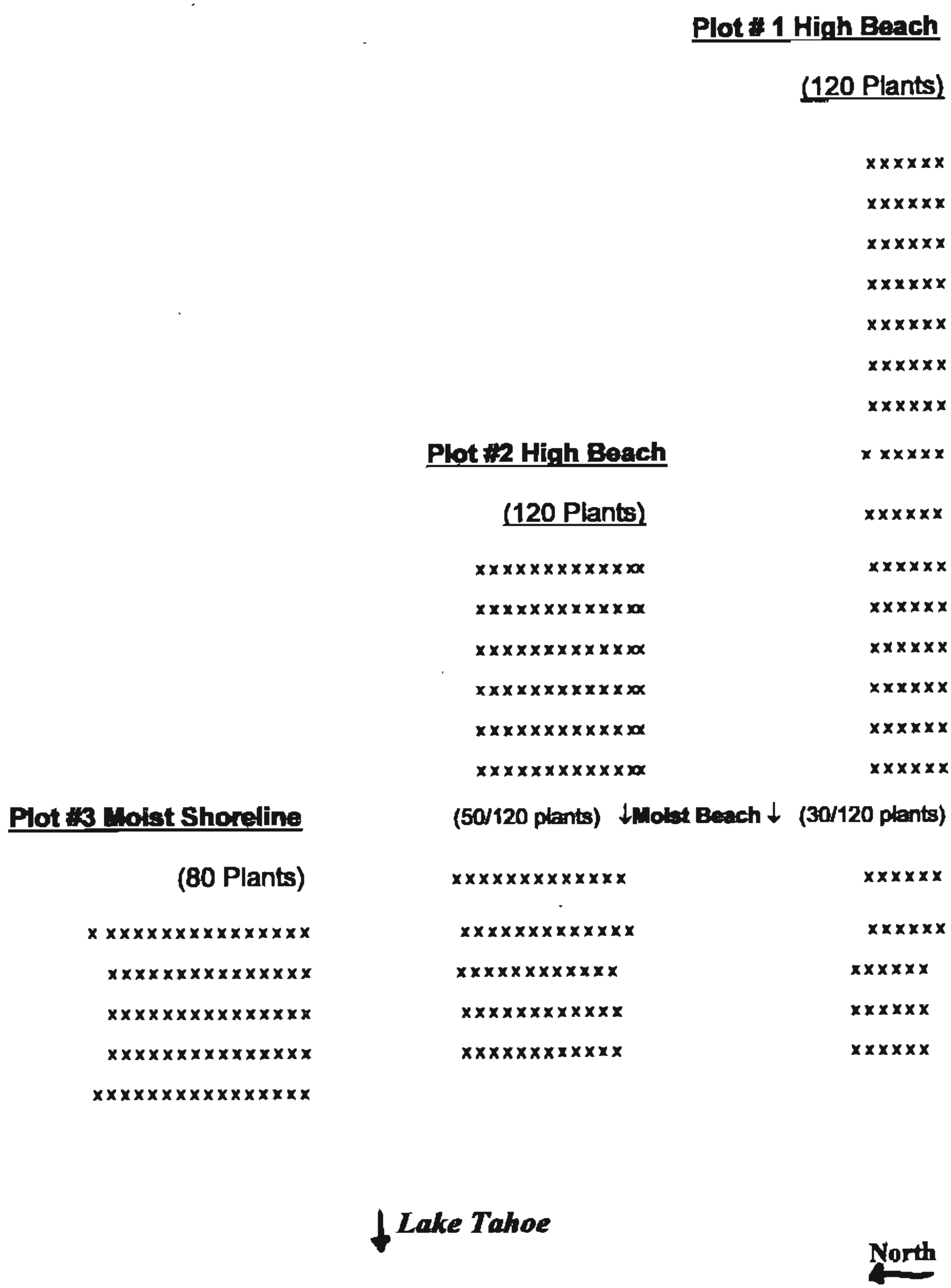
A2. Taylor Creek, Baldwin Beach (USFS)
Site Map 2003 Pilot Project
(Total Plants = 431)



A3. Zephyr (USFS)
Site Map 2003 Pilot Project
(Total Plants = 286)



A4. Sand Harbor (NDSP)
Site Map 2003 Pilot Project
(Total Plants = 320)



Appendix B. Protocols

Appendix B. Planting Protocol

1) Columns are perpendicular to shore, 1 meter apart. Rows are parallel to shore, ½ meter apart. Rows and columns may vary according to topography. Plant in columns only for minimum disturbance.

2) Flag color indicates the seed source population. Rubberband colors indicate estimated degree of vigor.

3) Gather the required plants from the tube racks and distribute them to the right of the appropriate flag. Degree of vigor (designated by rubberband color) chosen randomly.

4) Designate waterer. This person will prime the planting area with water. This watering prevents the caving in of loose soil. Each plot will be planted consistently, according to position from flag (i.e. all to the left of flag or all in front of flag). Group decides on positioning.

5) One person completes planting, one column at a time. Begin by attaching the seed source flag, for each plant, to the appropriate pinflag. Carefully, slice open plants in the stewy tubes with a boxcutter.

6) With a long-nosed shovel, make an opening parallel to the row, at least as deep as the tube. Don't dig, just push open a slot.

7) Gently, remove the TYC from the tube through the opening. Palm should support the sand-root mass, as it slides out. Do your best to keep roots protruding from the bottom of the tube intact.

8) If rootbound, loosen the roots gently with fingers. Less vigorous root systems require less manipulating.

9) Orient sand-root mass vertically in slot. Plant should be at or slightly below sand surface level.

10) Remove shovel and allow sand to relax around the mass. To completely enclose the hole, use pressure from your hand. Make sure no underground roots are exposed.

11) After an entire column is planted, designated waterer gives each plant water around its entire base (8" diameter). Amount of water will vary, due to the water table and the elevation of Lake Tahoe. Plant should not sink into a deep bowl. Waterer is responsible for ensuring that each of the plants in the plot has been sufficiently watered.

Appendix B. Mapping and QAQC Protocol

1) Arrange the necessary "project datasheets" (those with the dark circles) so that the primary sheet (the A-J, 1-25 sheet) is at the bottom left (so that position A1 is always left and lakeside when you are looking inland). Other sheets can be arranged, lettered (columns K, L, ...), and numbered (26, 27....) to accommodate any size plot.

2) Starting in column A, one person will call out the row of the first plant from lakeside and gives the necessary codes that apply to that plant. The code is S-P-V, which is population source # (1-8), phenology (V= vegetative, F=flower, Fr= fruit, S=senescent) and vigor (H,M,L).

3) After the code is recorded (e.g. "2-F-H") and called back by a second person (for confirmation), a meter stick is used to measure the short diameter (d1) and the long diameter (d2) of the plant, in cm. These are also recorded on the datasheet after the code ("2-F-H/5+7").

4) When the entire plot is mapped and confirmed, use a separate "plot map" to sketch the positions of prominent boulders, logs, shrubs, rush stands, etc.). Remove baseline tape and stakes.

5) Keep the project datasheets and plot maps in a notebook and make copies as soon as possible.

6) Use a light leaf rake to obliterate footprints, mounds, etc. from all aisles, buffer zones inside fence, and the 1 meter strip around the outside of the fence.

7) Take photos from permanent stake A1 looking towards J 25, stake J1 looking towards A 25, etc.

8) Signage should be in place.

Appendix B. Disturbance Monitoring Protocol

1) Using a copy of the plot maps for a site (now dated for this monitoring day), sketch in and label the following disturbance features that have appeared since last monitoring:

a) footprints/body impressions in the plot
b) footprints outside of the plot (up to 1 m away)
c) animal prints in the plot, especially dogs, geese
d) trash in the plot or along fence
e) windblown or water transported natural debris (if significant and obvious)
f) any acts of vandalism, especially those affecting TYC plants or the fence/signs

2) Assign a relative impact level (low, medium, high) to any of the above. Impact level is judged by effects on TYC plants, sand surface topography, extent of plot affected).

3) Record the number and positions of TYC plants that have been impacted (using the column and row numbers), and assigning a status to each:

a) obviously missing
b) obviously dead, but still in place
c) injured (stems, flowers broken off)
d) displaced but still rooted in place

4) Photograph the worst damage if it can illustrate the disturbance problem.

5) Repair damage to plants, if possible (if still rooted), making a note of what was done. Remove any detached or uprooted "carcasses" from plot and put in a paper bag. Remove any trash, but not natural materials that have come in.

6) Rake aisles, buffers and outside fence to smooth sand surface and obliterate the signs of disturbance.

7) Report all disturbances, especially the number of lost plants, to Alison Stanton.

Appendix B. Water Relations Monitoring Protocol

1) Water relations monitoring of outplanted TYC in the pilot project will be done twice during the 2003 growing season; once in early June (14 days after outplanting) and in early to mid September.

2) The monitoring days should be clustered (e.g. all sites on consecutive days), and seasonally "typical", meaning clear and sunny, warm, and not within 5 days after a stormfront has passed (and dropped wetting ppt).

3) Each site will have xylem water potentials measured at two times during the day:

predawn (5-6 am) – before direct light in the sky

midday (2-4 pm) – period with warmest temps, lowest humidity

4) Sites: Avalanche, Taylor Creek, Zephyr Cove (produces a west to east gradient)

5) At each sites, two microsites:

near the lake, low elevation (e.g. bottom 3-4 rows)

upper beach near stabilized sands, high elevation (e.g. upper 3-4 rows)

6) At a site, for each time of day and each microsite, 4 stems of outplanted TYC from 4 individuals will be used.

7) Individuals will be selected based on position, their good general appearance, and with a size sufficient to allow one stem to be excised without significant harm.

8) Excise and measure stems one at a time, cutting with a sharp, new razor blade straight across. Immediately insert it into the bomb chuck, tighten, and put into bomb lined with a moist paper towel.

9) The first measurement of a time period should be done slowly to not overshoot the endpoint. Record on datasheet in bars (e.g. 15.2), along with plant # (e.g. B4), the stem features, and the quality of the endpoint reading.

10) Excise another replicate stem and make measurement.

Appendix B. Demographic Monitoring Protocol

1) Demographic monitoring of outplanted TYC in the pilot project will be done five times during the 2003 growing season, along with disturbance monitoring on those dates and one additional date, July 7:

Demo + Disturb	June 2-3
Demo + Disturb	July 2+3
Disturb	July 7
Demo + Disturb	Aug 4 + 5
Demo + Disturb	Sept 1+2
Demo + Disturb	Oct 1+2

2) Two people are needed per site, on as a plot reader and one as a data writer.

3) Materials needed:

copy of plot map (for disturbance monitoring)
copy of previous monitoring datasheet or project datasheet
new monitoring datasheet
2 50 m tapes
2 metal stakes
light leaf rake
meter stick w/cm increments (last date only)
pencil
clipboard
camera

4) Approach plot from water's edge, below baseline. Go to the A1 corner and get oriented using the plot map and previous monitoring datasheet. Use the twistums on the fence to make sure you can identify rows.

5) Stretch a tape from the A1 permanent stake to the J1 permanent stake (and beyond if needed).

6) Do Disturbance monitoring (see protocol).

7) To do Demographic Monitoring, start from the A column, fix a second tape at the baseline with a stake and stretch it up the center of the column to confirm positions and identities of plants. They should be on 0.5 m centers and conform to what was previously mapped.

Appendix B. Demographic Monitoring Protocol (cont'd)

8) Plot reader finds the first plant up from the baseline and reads its position (e.g. A5) and status (live, dead, senescent, flowering, etc.). If the writer, looking at the previous datasheet, thinks a plant is missing, (e.g. there was a live plant at A3 on the last monitoring day), then the reader must search and confirm it is missing. An X is marked on position A3 if missing. Otherwise (i.e. if A5 is the first plant in the column) then the correct symbols are recorded in position A5 on the datasheet.

9) Move up the column, reading /confirming as you go.

10) When a column is finished, move one to the right and continue until all plants had been assigned a fate.
If you suspect that some natural TYC plants have invaded the plot, make a sketch map and notes.

11) On the last monitoring date (peak reproduction), the short diameter and long diameter of each plant will be measured to the nearest cm and recorded.

12) Collect and store all datasheets (all labeled, with dates, names, etc.) in a notebook. Make copies (which will be used on the next date) and keep originals in a safe place.

13) Rake aisles, buffers and outside fence to smooth sand surface and obliterate the signs of disturbance.

Appendix C. Sample Datasheets

TYC Water Relations Monitoring

Site Sand Harbor who AES date 4/27/03

Notes on recent weather beautiful: sunny all week

Predawn measurements (5-6 am)

Actual time of first measurement 0700 PDS sky conditions clear
Air temperature @ + 10 cm above soil surface 45 ~~C~~ F wind none
Soil temperature @ - 10 cm below surface ______ C

microhabitat	plant ID	stem sample descrip	bomb P (bars to 0.2)	endpoint quality
water edge	1) F8	grn, some yellowing, dehisced open fruit	4.8	good
	2) D15	grn c fruit only	2.4	good
finish →	3) E16	grn fruit & flwrs	2.0	
	4)			
high beach start	1) C11	lush grn with flwrs	3.8	good
	2) B10	grn some brwn lvs.	4.2	good
	3) A11	a little purple - thick	5.6	good
	4) C7	lush grn with fruit & flwrs	2.4	good

Midday measurements (2-4 pm)

Actual time of first measurement 2:45 PDS sky conditions clear
Air temperature @ + 10 cm above soil surface 82 ~~C~~ F wind none
Soil temperature @ - 10 cm below surface ______ C

microhabitat	plant ID	stem sample descrip	bomb P (bars to 0.2)	endpoint quality
water edge	1) C7	grn flwrs & beginning fruits	7.8	good
	2) F8	grn some yellow flwrs & dehisced fruit	9.8	good
	3) E9	grn flwrs	12.4.	good
	4) A7	grn flwrs, fruits	9.8	good
high beach first	1) D20	grn c flwrs	9.8	good
	2) B20	grn c flwrs	10.2	good
	3) C18	grn slightly purple c fruits	9.8	good
	4) A14	grn old flwrs in shade of fence	6.6	good

D15 grn c fruit 7.8 ok

TYC Pilot Reintroduction Project

site TAYLOR - PLOT 2 recorded by AS / BP # maps for this ~~site~~ plot 2 date 5/19/2003

gps: A1 38 56.473N 120 03.580W J1 38 56.472N 120 03.585W notes LOW BERM - TROUGH - LOW BEACH - BENCH

	A	B	C	D	E	F	G	H	I	J	
25	5SM	7SL	3FM	2SM	3FH	4SL	4VL	5VL	6VM	4VM	Inland
24	6VM	3SM	3FM	7FH	2FM	4VH	5VM	5FM	7VM	5VM	
23	2SM	3FH	6FM	4FH	7FH	3SL	5VH	5FM	5FM	7FH	
22											
21											
20											
19											
18											
17	7SL	2SM	2VL	4VH	4VM	6FM	4FH	5VM	5FH	6FH	
16	5SL	4VH	5FM	2SM	4SL	4VM	3FH	4VH	2VM	7VM	
15	6VM	2SM	5VM	3FH	7FH	2VM	3SL	4FH	5VH	3VH	
14											
13											
12											
11											
10											
9											
8											
7											
6	6SM	3FH	4FH	4VH	4VH	4VH	4VH	2FM	3VH	2VL	
5	5VM	3VH	4VH	3VH	3SL	7SM	3VH	2FL	6VL	4VM	
4	4VH	2VM	3FH	7FH	4VH	5VM	5VM	4FH	6SH	2VM	
3	4FH	5SM	6SL	4FH	3FH	2SL	2VM	4FH	3VL	5FM	
2	2VM	2VM	2SL	3SL	5SL	5SM	4FH	2SL	3SM	4VH	
1	6FH	4FM	6SM	2FH	4FH	4FH	4FH	7SL	7FM	6SL	water

Appendix D. Summary Data Tables

Avalanche

All Rows

Plot # 1	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
June	35	29	127	3	8	35	194	81.9	14.9
July	27	15	146	0	12	37	188	79.3	8.0
August	79	15	101	0	1	41	195	82.3	7.7
September	20	110	66	1	1	39	197	83.1	55.8
October	0	112	46	38	1	40	196	82.7	57.1
								81.9	**28.7**

Shoreline rows (1-4)

Plot #1	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
June	1	0	6	0	0	33	7	17.5	0.0
July	0	0	10	0	0	30	10	25.0	0.0
August	2	0	8	0	0	30	10	25.0	0.0
September	3	4	5	0	0	28	12	30.0	33.3
October	0	7	5	0	0	28	12	30.0	58.3

Low Beach Rows 5-24)

Plot #1	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
June	34	29	121	3	8	2	187	94.9	15.5
July	27	15	136	0	12	7	178	90.4	8.4
August	77	15	93	0	1	11	185	93.9	8.1
September	17	106	61	1	1	11	185	93.9	57.3
October	0	105	41	38	1	12	184	93.4	57.1

All Rows

Plot #2	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
June	8	11	30	9	2	0	58	96.7	19.0
July	9	7	42	0	2	0	58	96.7	12.1
August	19	10	29	0	2	0	58	96.7	17.2
September	13	33	10	1	1	2	57	95.0	57.9
October	0	44	11	3	0	2	58	96.7	75.9
								96.3	**36.4**

Plot 1and Plot2

Combined	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
June	43	40	157	12	10	35	252	84.8	15.9
July	36	22	188	0	14	37	246	82.8	8.9
August	98	25	130	0	3	41	253	85.2	9.9
September	33	143	76	2	2	41	254	85.5	56.3
October	0	156	57	41	1	42	254	85.5	61.4
								84.8	**30.5**

All Low BeachPlot 1 5-24 and Plot 2

Combined	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
June	42	40	151	12	10	2	245	82.5	16.3
July	36	22	178	0	14	7	236	79.5	9.3
August	96	25	122	0	3	11	243	81.8	10.3
September	30	139	71	2	2	13	242	81.5	57.4
October	0	149	52	41	1	14	242	81.5	61.6
								81.3	**31.0**

Taylor

All Rows

Plot # 1	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
2-Jun	15	1	24	12	0	7	52	86.7	1.9
16-Jun	1	9	15	6	0	28	31	51.7	29.0
1-Jul	2	1	8	3	0	43	14	23.3	7.1
1-Aug	3	10	4	2	0	39	19	31.7	52.6
2-Sep	0	17	0	0	0	41	17	28.3	100.0
1-Oct	1	17	8	0	0	33	26	43.3	65.4
30-Oct	0	3	7	15	0	34	25	41.7	12.0

Plot # 2	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
2-Jun	23	0	161	39	2	18	223	91.8	0.0
16-Jun	30	21	126	38	4	24	215	88.5	9.8
1-Jul	84	5	82	33	0	39	204	84.0	2.5
1-Aug	6	116	52	9	7	51	183	75.3	63.4
2-Sep	1	126	43	3	0	68	173	71.2	72.8
1-Oct	0	98	40	37	0	68	175	72.0	56.0
30-Oct	0	41	56	72	0	74	169	69.5	24.3

Plot # 3	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
2-Jun	22	20	30	21	6	9	93	86.1	21.5
16-Jun	2	31	36	19	1	18	88	81.5	35.2
1-Jul	29	9	31	10	3	26	79	73.1	11.4
1-Aug	0	39	21	5	1	41	65	60.2	60.0
2-Sep	2	31	24	3	0	47	60	55.6	51.7
1-Oct	0	23	24	14	0	46	61	56.5	37.7
30-Oct	0	12	26	23	1	45	61	56.5	19.7

Plot # 4	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
2-Jun	25	10	17	23	9	6	75	83.3	13.3
16-Jun	5	25	26	11	3	20	67	74.4	37.3
1-Jul	22	5	37	2	1	23	66	73.3	7.6
1-Aug	0	43	20	0	0	26	63	70.0	68.3
2-Sep	5	42	18	1	0	24	66	73.3	63.6
1-Oct	0	40	18	3	0	29	61	67.8	65.6
30-Oct	0	10	16	30	3	31	56	62.2	17.9

Plot # 5	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
2-Jun	10	2	11	12	1	4	35	87.5	5.7
16-Jun	0	0	2	13	15	10	15	37.5	0.0
1-Jul	0	0	1	0	2	37	1	2.5	0.0
1-Aug	0	0	0	0	1	39	0	0.0	0.0
2-Sep	0	0	0	0	0	40	0	0.0	0.0
1-Oct	0	0	0	0	0	40	0	0.0	0.0

Plots1-4, no 5

Combined	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
2-Jun	85	31	232	95	17	40	443	88.4	7.0
16-Jun	38	86	203	74	8	90	401	80.0	21.4
1-Jul	137	20	158	48	4	131	363	72.5	5.5
1-Aug	9	208	97	16	8	157	330	65.9	63.0
2-Sep	8	216	85	7	0	180	316	63.1	68.4

1-Oct	1	178	90	54	0	176	323	64.5	55.1
30-Oct	0	66	105	140	4	184	311	62.1	21.2

All Plots **Combined**	**Flower**	**Fruit**	**Vegetative**	**Senescent**	**Dead**	**Missing**	**#alive**	**% alive**	**% Repro**
2-Jun	95	33	243	107	18	44	478	88.4	6.9
16-Jun	38	86	205	87	23	100	416	76.9	20.7
1-Jul	137	20	159	48	6	168	364	67.3	5.5
1-Aug	9	208	97	16	9	196	330	61.0	63.0
2-Sep	8	216	85	7	0	220	316	58.4	68.4
1-Oct	1	178	90	54	0	216	323	59.7	55.1

Zephyr
All Rows

Plot # 1	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
2-Jun	10	19	92	12	0	22	133	85.3	14.3
16-Jun	16	29	43	11	4	53	99	63.5	29.3
1-Jul	25	20	54	1	0	55	100	64.1	20.0
1-Aug	9	75	12	0	0	60	96	61.5	78.1
2-Sep	1	84	12	1	0	57	98	62.8	85.7
1-Oct	0	87	21	0	0	47	108	69.2	80.6
30-Oct	0	37	20	50	1	47	107	68.6	34.6

Shoreline rows (1-4)

Plot #1	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
2-Jun	0	2	28	1	0	21	31	59.6	6.5
16-Jun	0	1	2	0	0	49	3	5.8	33.3
1-Jul	0	0	10	0	0	42	10	19.2	0.0
1-Aug	0	0	6	0	0	46	6	11.5	0.0
2-Sep	1	0	8	0	0	43	9	17.3	0.0
1-Oct	0	1	16	0	0	35	17	32.7	5.9
30-Oct	0	1	14	1	1	35	16	30.8	6.3

Low Beach rows (5-12)

Plot #1	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
2-Jun	10	17	64	11	0	1	102	98.1	16.7
16-Jun	16	28	41	11	4	4	96	92.3	29.2
1-Jul	25	20	44	1	0	14	90	86.5	22.2
1-Aug	9	75	6	0	0	14	90	86.5	83.3
2-Sep	0	84	4	1	0	14	89	85.6	94.4
1-Oct	0	86	5	0	0	12	91	87.5	94.5
30-Oct	0	36	6	49	0	12	91	87.5	39.6

All Rows

Plot # 2	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
2-Jun	23	32	60	7	2	6	122	93.8	26.2
16-Jun	0	30	25	55	2	16	110	84.6	27.3
1-Jul	1	28	53	37	3	28	119	91.5	23.5
1-Aug	4	18	36	20	1	50	78	60.0	23.1
2-Sep	5	22	39	3	1	60	69	53.1	31.9
1-Oct	2	15	57	0	0	54	74	56.9	20.3
30-Oct	0	17	56	0	0	56	73	56.2	23.3

both plots

Combined	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
2-Jun	33	51	152	19	2	28	255	88.9	20.0
16-Jun	16	59	68	66	6	69	209	72.8	28.2
1-Jul	26	48	107	38	3	83	219	76.3	21.9
1-Aug	13	93	48	20	1	110	174	60.6	53.4
2-Sep	6	106	51	4	1	117	167	58.2	63.5
1-Oct	2	102	78	0	0	101	182	63.4	56.0
30-Oct	0	54	76	50	1	103	180	62.7	30.0

high beach

plot1 5-12+ plo	**Flower**	**Fruit**	**Vegetative**	**Senescent**	**Dead**	**Missing**	**#alive**	**% alive**	**% Repro**
2-Jun	33	49	124	18	2	7	224	78.0	21.9
16-Jun	16	58	66	66	6	20	206	71.8	28.2
1-Jul	26	48	97	38	3	42	209	72.8	23.0
1-Aug	13	93	42	20	1	64	168	58.5	55.4
2-Sep	5	106	43	4	1	74	158	55.1	67.1
1-Oct	2	101	62	0	0	66	165	57.5	61.2
30-Oct	0	53	62	49	0	68	164	57.1	32.3

Sand Harbor

All Rows

Plot # 1	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
June	5	0	17	54	27	14	76	65.0	0.0
July	1	0	24	22	33	37	47	40.2	0.0
August	11	0	35	0	26	45	46	39.3	0.0
September	7	25	20	0	3	62	52	44.4	48.1
October	8	24	9	7	3	66	48	41.0	50.0

Shoreline rows (1-4)

Plot #1	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
June	1	0	9	37	0	90	47	34.1	0.0
July	0	0	4	20	1	113	24	17.4	0.0
August	0	0	1	0	0	137	1	0.7	0.0
September	**0**	**0**	**1**	**0**	**0**	**137**	1	0.7	0.0
October	0	1	3	0	0	134	4	2.9	25.0

Low Beach plot 1 5-20, plot 2 5-10,

Plot #1	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
June	8	0	36	61	37	10	105	69.5	0.0
July	4	4	38	26	34	45	72	47.7	5.6
August	20	1	49	6	26	49	76	50.3	1.3
September	15	34	29	0	3	70	**78**	51.7	43.6
October	11	39	16	10	4	72	76	50.3	51.3

All Rows

Plot #2	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
June	4	0	20	25	8	46	49	47.6	0.0
July	3	0	8	24	2	66	35	34.0	0.0
August	9	1	14	6	0	73	30	29.1	3.3
September	8	9	9	0	0	77	26	25.2	34.6
October	3	15	6	3	1	75	27	26.2	55.6

Plot #3	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
June	0	0	8	19	2	40	27	39.1	0.0
July	0	4	10	0	0	55	14	20.3	28.6
August	0	0	1	0	0	68	1	1.4	0.0
September	0	0	1	0	0	68	1	1.4	0.0
October	0	1	4	0	0	65	5	7.2	20.0

all

Combined	Flower	Fruit	Vegetative	Senescent	Dead	Missing	#alive	% alive	% Repro
June	9	0	45	98	37	100	152	52.6	0.0
July	4	4	42	46	35	158	96	33.2	4.2
August	20	1	50	6	26	186	77	26.6	1.3
September	15	34	30	0	3	207	79	27.3	43.0
October	11	40	19	10	4	206	80	27.7	50.0